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Additional Distributional Records of *Ambystoma laterale*, *A. jeffersonianum* (Amphibia: Caudata) and Their Unisexual Kleptogens in Northeastern North America

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ABSTRACT

Several species of mole salamanders in the genus *Ambystoma* are targeted by various state, provincial, and federal agencies for conservation. These salamanders have specific wetland and forested upland habitat requirements that render them vulnerable to environmental alteration. The blue-spotted salamander, *Ambystoma laterale* (LL) and the Jefferson salamander, *A. jeffersonianum* (JJ) have both been listed for protection in various parts of their ranges, but the identification of these salamanders is confusing because they often coexist with unisexual individuals that are mostly polyploid and use the sexual species as sperm donors. We used isozyme electrophoresis, blood erythrocytes, and chromosome counts in a continued effort to identify sexual and unisexual individuals in eastern North America. We examined 1377 salamanders from 118 sites in Connecticut, Massachusetts, New Jersey, New York, Pennsylvania, and Virginia. Most Pennsylvania salamanders were *A. jeffersonianum* (JJ) but *A. laterale* (LL), previously unknown from Pennsylvania, were found in that state. The two sexual species were never found together. We found diploid (LJ), triploid (LLJ; LJJ), and tetraploid (LLLJ; LJJJ; LLJJ) unisexuals. At most collecting sites, unisexuals were more numerous than sexual individuals. The association of sexual and unisexual individuals support a kleptogenic reproductive system in which the unisexuals steal genomes from their sympatric sexual sperm donors.

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TABLE 1
Conservation status of *Ambystoma laterale* and *A. jeffersonianum* populations in Connecticut, Massachusetts, New Jersey, New York, Pennsylvania, and Rhode Island
E=endangered, T=threatened, SC=special concern.

Genetic composition of individual breeding site	CT	MA	NJ	NY	PA	RI
<i>A. jeffersonianum</i> complex	SC	SC	SC	SC	No conservation status (not considered native to PA) ^b	Not known from RI ^a
<i>A. jeffersonianum</i>	Not known from CT ^a	Not known from MA ^a	Not known from NJ ^a	SC	No conservation status in PA	Not known from RI ^a
<i>A. laterale</i> complex	SC	SC	E	SC	No conservation status (not considered native to PA) ^c	Not known from RI ^a
<i>A. laterale</i>	T	SC ^d	E	SC ^d	No conservation status (not considered native to PA)	Extirpated ^e

^a Bogart and Klemens, 1997; Bogart and Klemens, present study.
^b This study confirms the presence of unisexuals associated with *A. jeffersonianum* in northeastern PA (Monroe Co.).
^c This study confirms the presence of *A. laterale* and associated unisexuals in northwestern PA (McKean Co.) and northeastern PA (Northampton Co.).
^d MA and NY do not recognize the unique conservation status of pure diploid *A. laterale* populations in southeastern MA and eastern Long Island, NY.
^e Pure diploid *A. laterale* was reported from the Pawtucket area of RI by Drowne (1905). These populations are considered extirpated (C. Raithel, personal commun.).

INTRODUCTION

There are about 30 living species of mole salamanders in the North American family Ambystomatidae (Petranka, 1998). Most species occur in the continental United States, but there has been a Mexican invasion of the *Ambystoma tigrinum* complex (Shaffer and McKnight, 1996) and some species have northern ranges that include parts of Canada (Petranka, 1998). Mole salamanders often have specific habitat requirements that make them vulnerable to anthropogenic environmental alterations and most species are listed as candidates for conservation in some or all parts of their range. The blue-spotted salamander, *A. laterale* and the Jefferson salamander, *A. jeffersonianum* present problems for conservationists because they both coexist with unisexual individuals that normally do not have a conservation status (Kraus, 1995). Connecticut lists *Ambystoma jeffersonianum* complex and *A. laterale* complex as state species of special concern because it is difficult to distinguish the unisexuals from the sexual species (Klemens, 2000). Connecticut also lists the pure diploid populations of *A. laterale* in the eastern portion of the state as a threatened

species (Klemens, 2000). Connecticut is the only state in the northeastern United States that has differentially protected the pure diploid *A. laterale* populations that also occur in southeastern Massachusetts and on the eastern tip of Long Island, New York. The conservation status of these salamanders does vary over their ranges (table 1).
We (Bogart and Klemens, 1997) provided a detailed historical perspective on the unisexual salamanders and their associated sperm donor species. We also identified and characterized populations of *A. laterale*, *A. jeffersonianum*, and their unisexual associates in the New England states and parts of New York. Based on 1002 individuals from 106 sites, we found the unisexuals to be common and widespread. The majority of the populations that contained either *A. laterale* or *A. jeffersonianum* also contained unisexual individuals. Seventy percent of the salamanders in that study were found to be unisexuals. Ten of the 106 sites were found to contain only *A. laterale*, but six of these sites were represented by one or two individuals. Samples obtained from two of the three sites where only *A. jeffersonianum* were found consisted of single specimens and only three individuals were sampled from the other

A. jeffersonianum site. Diploid, triploid, and tetraploid unisexuals were found throughout New England. Most unisexual salamanders were polyploid, but, because diploid unisexuals were found at 21 scattered sites, ploidy is not a dependable distinguishing character to separate unisexual and sexual individuals without corroborative data. Unisexual salamanders are expected to be female, but rare "unisexual" males were also found; thus, knowing that an individual is a male does not always confirm a sexual species identification.

Clarification of the evolution of unisexual salamanders and the reproductive mode of these females has been further elucidated using gene sequences and microsatellite DNA loci (Bogart, 2003; Bogart et al., 2007). Mitochondrial DNA analysis show that the unisexuals arose about three to four million years ago from a hybridization event involving a female that shared its most recent common ancestor to individuals of *A. barbouri* from Kentucky. *Ambystoma barbouri* has a current, restricted range in Ohio, Kentucky, and Tennessee and is a recently uncovered (Kraus and Petranksa, 1989) sister species (Petranksa, 1998; Niedzwiecki, 2005) to *A. texanum*. Subsequently, unisexuals rapidly dispersed in eastern North America using a reproductive mode that appears to be unique (Bogart et al., 2007). All unisexuals maintain a similar *A. barbouri*-like mitochondrial DNA but can incorporate and exchange nuclear genomes with sympatric males. Because unisexual females can steal genomes from a variety of sympatric males, the suggested reproductive mode is kleptogenesis (Bogart et al., 2007) and has given rise to at least 20 nuclear genomic combinations, or kleptogens, that are diploid, triploid, tetraploid, and even pentaploid (Bogart, 2003).

To expand the coverage of populations and to include more of the range of these salamanders in New Jersey, New York, and Pennsylvania the present study used isozyme electrophoresis to identify additional individuals of sexual *A. laterale*, *A. jeffersonianum*, and unisexual kleptogens that use these species as sperm donors. We also sampled new populations in New England and included a few samples from populations that were

previously sampled by Bogart and Klemens (1997) to increase the sample size and to confirm our findings in previously sampled populations. Accurate data on the distribution of this salamander complex should assist conservation efforts to protect these salamanders. Such data are also necessary to explore the evolutionary significance of kleptogenesis.

MATERIALS AND METHODS

Methods were mostly identical to those used by Bogart and Klemens (1997). Salamanders were collected with dip nets and minnow traps from breeding ponds early in the spring, while crossing roads at night during spring rains, or from under rocks and logs during the day from March through October. Egg mass surveys (Bogart, 1982) were not done because data obtained from such surveys may not provide a random sample of the population as most eggs laid by unisexual females do not result in hatched larvae and many hatched larvae do not survive to transformation. In some cases, when adults were not collected from a breeding pond, advanced larvae were randomly collected using a dip net. They were maintained through transformation in the laboratory and the juveniles were considered to be representative of the population or site. Our present study is a continuation of the collections done for our previous study (Bogart and Klemens, 1997). Collections for the present study were conducted over a nine-year period, from 1996 to 2004. Collections were concentrated in New York, New Jersey, and Pennsylvania, but collections also included sites in Connecticut, Massachusetts, and Virginia. Salamanders (1377 individuals) from 118 sites (appendix 1) were collected and shipped to Guelph for identification. Salamanders were injected with colchicine (0.25 to 0.75 ml of a 0.1 mg/ml solution) two days before they were killed by prolonged anesthesia in a 7% solution of buffered (pH 7.0) tricaine methane sulfonate (MS222). The heart was exposed and blood was collected from the conus arteriosus in heparinized microhematocrit tubes for ploidy determination. The intestine, including the cloaca, was dissected from each individual, immersed in de-ionized water for 15 min, fixed in 3:1

TABLE 2
Presumptive structural gene loci examined in *Ambystoma*

Locus (abbreviation) ^a	EC No. ^b	Tissue ^c	Gel ^d
Aspartate aminotransferase (Aat-1 ^a) (= Got-1 ^c)	2.6.1.1	HMS	2
Aat-2 ^a (= Got-2 ^c)	2.6.1.1	L	2
Isocitrate dehydrogenase (Idh-1 ^a) (= ICD-1)	1.1.1.42	HMS	1
Lactate dehydrogenase (Ldh-1 ^a)	1.1.1.27	HMS	1
Ldh-2	1.1.1.27	HMS	1
Malate dehydrogenase (Mdh-1 ^a)	1.1.1.37	HMS	1
Mannose-6-phosphate isomerase (Mpi1) (= PGDH)	5.3.1.8	L	1
Phosphoglucose isomerase (Pgi) (= GPI)	5.3.1.9	HMS	2
Phosphoglucomutase (Pgm-1)	2.7.5.1	L	1
Pgm-2	2.7.5.1	L	1
Superoxide dismutase (Sod-1 ^a)	1.15.1.1	L	1

^a Loci that were found to be most useful for distinguishing *A. laterale*, *A. jeffersonianum*, and associated unisexuals. The abbreviations and synonyms are those used in this and most previous electrophoretic studies of vertebrates.

^b Standardized enzyme-numbering system established by the nomenclature committee of the International Union of Biochemistry (IUBC, 1984).

^c Tissues used to resolve the enzyme systems were liver (L) or a combination of heart, skeletal muscle, and spleen (HMS).

^d The electrophoretic conditions for the gels were: (1) amine-citrate, gel, and tray buffer adjusted to pH 6.5 (Clayton and Tretiak, 1972) run for 3 hr at 250 volts; (2) tris-citrate, gel buffer pH 6.7, and tray buffer pH 6.3 (Selander et al., 1971) run 3 to 4 hr at 150 volts.

^e Got (glutamate oxaloaceto-transaminase) is a synonym of aspartate aminotransferase (Aat). Previous studies on *Ambystoma* used this earlier enzyme designation (Got).

ethanol:acetic acid in 1.5 ml Eppendorf microtubes, and stored at -20°C for chromosome analyses. Tissues required for isozyme electrophoresis were liver and a combination of the heart, skeletal muscle, and spleen. The tissues were removed from freshly killed salamanders and stored with an equal volume of de-ionized water in 1.5 ml Eppendorf microtubes in an ultracold freezer (-80°C). After the tissues were removed, the specimens were preserved and deposited at the American Museum of Natural History (AMNH) (appendix 1).

ISOZYME ELECTROPHORESIS

Just prior to electrophoresis, the frozen tissues were ground using a sharp glass rod and spun for two minutes in a microcentrifuge. Filter-paper wicks were used to soak up a portion of the supernatant. The wicks were air dried on filter paper and inserted in starch gels. Horizontal starch-gel electrophoresis followed the procedures outlined by Selander et al. (1971), Bogart (1982), and Bogart and Klemens (1997). The buffer systems used were described by Selander et al. (1971) and Clayton and Tretiak (1972). Electrophoretic

loci for each enzyme system were numbered on the gel from the most anodally migrating locus. As in previous studies (Bogart et al., 1987; Bogart, 1989; Bogart and Klemens, 1997), alleles, or allozymes, were designated by their relative mobilities compared with the mobility of the most common allele in *A. laterale*, which was assigned a mobility of 100. We chose 11 of the 21 isozyme loci used by Bogart and Klemens (1997) in our present study. These loci proved to be most useful in identifying the sexual and unisexual individuals and included loci that demonstrated some homozygosity and reversals in our previous study. The loci that were assayed, buffer systems used, and the tissues examined are provided in table 2. Nine loci were dimeric enzymes that have been the most useful in visualizing staining intensities of the bands (dosage) for assessing genome composition in polyploid amphibians (Danzmann and Bogart, 1982). Lactate dehydrogenase is a tetrameric enzyme that also expresses differential staining of the heterotetrameric bands. The only monomeric enzyme, phosphoglucose mutase, often demonstrated different staining intensities for two loci (pgm-1; pgm-2) and was previously found to possess a few rare

heterozygotes at *pgm-2* in both sexual and unisexual salamanders (Lowcock and Bogart, 1989; Bogart and Klemens, 1997).

PLOIDY DETERMINATION

A small drop of blood from the hematocrit tube, taken from every individual, was mixed with an approximately equal amount of diluted (25 ml H₂O added to 100 ml) Hanks Balanced Salt Solution (Sigma) and photographed under phase-contrast and bright-field optics. The area of the blood cells was determined using a sonic digitizer on a rear-projection image of the negatives (Austin and Bogart, 1982). Six blood cells from each individual were measured to obtain an average erythrocyte area. During our investigation, a flow cytometric method (FCM) was found to be a more accurate method to determine ploidy and that method was introduced into our study in 2003 following the methods in Ramsden et al. (2006). Blood cell data provided the first clear indication of diploid and tetraploid unisexuals that may not be easily distinguished from triploids using only the electrophoretic patterns. Chromosome numbers were the absolute proof of ploidy in the salamanders and those data were required if a discrepancy existed between the electrophoretic genotype, blood cell area, or flow cytometric analyses. When necessary, chromosomes were obtained from the gut epithelial tissue using procedures outlined by Kezer and Sessions (1979) and Sessions (1982). Unstained, or conventionally stained, chromosomes of *A. laterale* and *A. jeffersonianum* are virtually indistinguishable (Taylor and Bogart, 1990), so the chromosomes obtained for most individuals and those observed in our previous study (Bogart and Klemens, 1997) could not be used to assign genomic constitution in the unisexuals. New chromosome methods that use fluorescent genomic probes and in situ hybridization clearly differentiate genomes of these two species in the unisexuals (Bi and Bogart, 2006). Microsatellite DNA loci that are amplified using primers developed for *A. jeffersonianum* (Julian et al., 2003) also proved to be an accurate method to determine ploidy and genomic constituents in unisexual individuals (Ramsden et al., 2006;

Bogart et al., 2007). Although we employed all of these methods to confirm ploidy in individual salamanders, the analyses of chromosome and microsatellite data from tissue collected from individuals in the present study will be the subject of future investigations that use these new techniques.

SITE DESIGNATION

Because we were interested in knowing the sexual and unisexual associations in breeding populations, we tried to analyze the individuals that were expected to breed in the same population. In most cases, salamanders could be assigned to distinct breeding ponds because they were collected as adults or larvae in a pond or as newly transformed juveniles at the edge of a pond. Sometimes, however, individuals were collected on roads or in upland wooded areas and could not be assigned to any particular breeding pond. Additionally, salamanders were found at different locations entering or leaving extensive swamps that may be partitioned into subpopulations. These factors required the use of site designations based on the assumption that individuals from the same site shared a potentially common breeding area. A site may or may not be equated with a breeding population. In some instances, we combined individuals into a single site even though they were collected from extensive, yet ecologically similar, and contiguous habitat. Some other collections that were close geographically were treated as separate sites if they were distinctly separated by habitat and, in some instances, elevation. Klemens (1993) found that topography plays an important role in determining wetland type, which in turn influences the distribution of the sexual species. This was most apparent in areas of close contact where breeding ponds of the two sexual species were separated by as little as 100 meters. We grouped our sites into drainage basins because Klemens (1978, 1993) demonstrated that there were significant differences in the herpetofauna of New England's drainage basins, as a result of post-Pleistocene dispersion of amphibians into the interior of New England.

We numbered the sites starting with 107 as a continuation of the 106 sites sampled in our

TABLE 3
Allele frequencies of diploid and polyploid salamanders for each of the genotypes in appendix 2

<i>A. laterale</i> (LL), <i>A. jeffersonianum</i> (JJ), diploid, triploid, and tetraploid unisexual genotypes ^b									
Locus ^a	LL	JJ	LJ	LLJ	LJJ	LLLJ	LJJJ	LLJJ	Mobility ^c
Aat-1	(308) ^d	(463)	(86)	(227)	(244)	(26)	(20)	(1)	
A	0.003	—	—	—	—	—	—	—	+110
B	0.997	—	0.500	0.659	0.335	0.750	0.250	0.500	+100
D	—	1.000	0.500	0.341	0.665	0.250	0.750	0.500	+79
Aat-2	(306)	(462)	(86)	(223)	(242)	(25)	(20)	(1)	
A	—	0.995	0.500	0.335	0.658	0.250	0.750	—	−180
B	0.995	0.003	0.500	0.665	0.324	0.750	0.250	1.000	−100
C	0.005	0.002	—	—	—	—	—	—	−50
Idh-1	(307)	(463)	(86)	(218)	(241)	(26)	(20)	(1)	
Q	0.003	0.002	0.006	—	0.004	—	—	—	+160
A	—	0.998	0.494	0.324	0.650	0.240	0.812	0.500	+142
B	0.995	—	0.500	0.676	0.346	0.760	0.188	0.500	+100
C	0.002	—	—	—	—	—	—	—	+50
Ldh-1	(302)	(462)	(85)	(220)	(243)	(26)	(20)	(1)	
A	0.003	0.002	—	0.003	—	—	—	—	+115
B	0.886	—	0.506	0.629	0.348	0.750	0.300	0.500	+100
C	0.002	0.998	0.494	0.327	0.652	0.250	0.700	0.500	+88
D	0.109	—	—	0.041	—	—	—	—	+78
Ldh-2	(306)	(463)	(83)	(222)	(243)	(26)	(20)	(1)	
Q	—	0.003	—	0.002	—	—	—	—	+160
A	0.230	—	0.060	0.123	0.027	0.038	0.012	—	+130
B	0.767	0.997	0.874	0.874	0.971	0.952	0.988	1.000	+100
C	0.003	—	0.066	0.002	0.001	0.010	—	—	+55
Mdh-1	(306)	(462)	(92)	(227)	(244)	(26)	(20)	(1)	
A	—	0.002	—	—	0.001	—	—	—	+200
B	—	0.948	0.500	0.352	0.664	0.250	0.750	0.500	+176
C	0.005	—	—	—	0.008	—	—	—	+135
D	0.995	0.050	0.500	0.648	0.326	0.750	0.250	0.500	+100
Mpi	(253)	(316)	(58)	(180)	(186)	(16)	(9)	(1)	
A	—	0.003	0.086	0.007	0.027	—	—	—	+140
B	—	0.967	0.509	0.320	0.640	0.344	0.778	—	+120
C	0.990	0.028	0.388	0.672	0.330	0.656	0.222	1.000	+100
D	0.010	0.002	0.017	—	0.004	—	—	—	+80
Pgi	(162)	(274)	(42)	(153)	(235)	(20)	(16)	(1)	
Q	—	0.005	—	0.002	—	—	—	—	+380
A	0.037	0.663	0.381	0.285	0.567	0.225	0.531	0.500	+325
B	0.009	0.316	0.083	0.102	0.074	0.150	0.094	—	+115
C	0.954	0.016	0.536	0.610	0.359	0.625	0.375	0.500	+100
Pgm-1	(306)	(453)	(85)	(216)	(235)	(25)	(19)	(1)	
A	0.015	0.001	—	0.002	—	—	—	—	+115
B	0.974	0.646	0.794	0.821	0.675	0.940	0.647	1.000	+100
C	0.011	0.346	0.206	0.176	0.325	0.060	0.353	—	+97
D	—	0.007	—	0.002	—	—	—	—	+82

TABLE 3
(Continued)

<i>A. laterale</i> (LL), <i>A. jeffersonianum</i> (JJ), diploid, triploid, and tetraploid unisexual genotypes ^b									
Locus ^a	LL	JJ	LJ	LLJ	LJJ	LLLJ	LJJJ	LLJJ	Mobility ^c
Pgm-2	(308)	(460)	(86)	(219)	(243)	(26)	(20)	(1)	
A	0.018	—	—	—	—	—	—	—	+120
B	0.980	1.000	1.000	1.000	0.999	1.000	1.000	1.000	+100
C	0.002	—	—	—	0.001	—	—	—	+75
Sod-1	(313)	(463)	(86)	(226)	(244)	(24)	(20)	(1)	
B	1.000	—	0.483	0.671	0.333	0.769	0.238	0.500	+100
D	—	1.000	0.517	0.329	0.667	0.231	0.762	0.500	+37

^a Isozyme loci used to identify the sexual and unisexual individuals. The allozymes or alleles that were observed are included under each locus.

^b Genomotypes are the nuclear genomes inferred by the alleles that are diagnostic for *Ambystoma laterale* (L) and *A. jeffersonianum* (J) found in each individual.

^c Mobilities are the relative mobilities on the gel compared with the most common allele found in *A. laterale* that is assigned a mobility of 100. Positive mobilities (+) indicate migration from the origin toward the anode and negative mobilities (–) indicate migration toward the cathode.

^d Sample sizes in parentheses (n) are the number of individuals of each genomotype that were examined and scored for each locus (from appendix 2).

earlier study (Bogart and Klemens, 1997). Eight sites that were re-sampled from our earlier study were not given new site numbers in the present study.

NOMENCLATURE

Identifying and naming unisexual individuals is a formidable challenge for taxonomists and conservationists. The unisexuals do have a hybrid nuclear genomic constitution, but they are not hybrids that have resulted from the crossing of *A. laterale* and *A. jeffersonianum* or any combination of the four species whose genomes might be found in a unisexual. Genomes are gained and lost by kleptogenesis, which is driven entirely by male sperm donors. In this flexible genetic system, a single unisexual female can produce offspring that have differing genotypes and genomes (Bogart et al., 1987, 2007) and, because of intergenomic interaction (Bi and Bogart, 2006; Bi et al., 2007), genomes found in offspring from a particular unisexual might evolve within that unisexual independently of male genomic contribution. Lowcock et al. (1987) suggested an informal descriptive system for unisexual *Ambystoma* that was used by Schultz (1969) to describe the genetic composition of hybridogenetic unisexual fish of the genus *Poeciliopsis*.

Letter designations for species that contributed genomes to unisexual *Ambystoma* have previously been used for convenience in a number of previous studies (Uzzell, 1964; Bogart et al., 1985, 1987; Bogart and Klemens, 1997): J for *A. jeffersonianum*, L for *A. laterale*, T for *A. texanum*, and Ti for *A. tigrinum*. The proposed name, for example, for a triploid unisexual that has a nuclear genome consisting of one *A. laterale* genome and two *A. jeffersonianum* genomes would be *Ambystoma laterale* – (2) *jeffersonianum*, or LJJ.

GENOTYPE AND GENOMOTYPE ANALYSIS

The unisexuals are mostly fixed heterozygotes for a number of isozyme alleles and their mode of reproduction is kleptogenesis. Therefore, population genetic models that are based on randomly interbreeding individuals do not apply to the unisexual kleptogens. When unisexuals occur in a population, they usually outnumber individuals of the sexual species (Bogart and Klemens, 1997). If the sample of individuals obtained from a site contained unisexuals, but neither *A. laterale* nor *A. jeffersonianum* individuals were found, we suspected that the sample was too small to have encountered these species. In order to obtain some measure of the influence, or the

TABLE 4
Ambystoma laterale (LL), *A. jeffersonianum* (JJ), and nuclear hybrid genomes found at each site

Site	(n) ^a	Males ^b	Genome ^c								%L ^d
			2n			3n		4n			
			LL	JJ	LJ	LLJ	LJJ	LLLJ	LLJJ	LJJJ	
2†	(35)	24	—	35	—	—	—	—	—	—	00.0
7†	(4)	0	—	—	—	1	—	3	—	—	73.3
18†	(5)	1	2	—	1	1	—	—	1	—	69.2
20†	(2)	0	—	—	—	—	2	—	—	—	33.3
28†	(1)	0	—	—	—	—	1	—	—	—	33.3
42†	(6)	0	1	—	—	5	—	—	—	—	70.6
60†	(2)	1	2	—	—	—	—	—	—	—	100.0
66†	(2)	1	2	—	—	—	—	—	—	—	100.0
107	(25)	3	9	—	4	7	—	5	—	—	76.1
108	(43)	11*	14	—	7	17	2	3	—	—	72.1
109	(9)	0	—	—	2	—	4	—	—	3	31.0
110	(31)	6	19	—	1	9	—	2	—	—	85.3
111	(26)	3	9	—	—	14	—	3	—	—	68.0
112	(5)	1*	—	—	1	4	—	—	—	—	64.3
113	(43)	4	6	—	18	17	1	1	—	—	64.2
114	(46)	20*	—	44	1	—	1	—	—	—	2.2
115	(3)	1	—	1	—	—	2	—	—	—	25.0
116	(1)	0	—	1	—	—	—	—	—	—	00.0
117	(9)	0	—	—	—	—	5	—	—	4	29.0
118	(16)	10	—	16	—	—	—	—	—	—	00.0
119	(7)	0	—	1	—	—	6	—	—	—	30.0
120	(14)	5	—	14	—	—	—	—	—	—	00.0
121	(20)	13	—	20	—	—	—	—	—	—	00.0
122	(7)	1*	—	—	4	—	3	—	—	—	41.2
123	(14)	1	—	1	11	1	1	—	—	—	46.7
124	(11)	0	—	1	3	—	7	—	—	—	34.5
125	(5)	0	—	2	2	1	—	—	—	—	36.4
126	(30)	3	—	5	5	—	18	—	—	2	31.2
127	(51)	23	41	—	—	10	—	—	—	—	91.1
128	(1)	0	—	—	—	1	—	—	—	—	66.7
129	(12)	2	8	—	—	4	—	—	—	—	85.7
130	(13)	1	—	6	1	—	6	—	—	—	21.9
131	(6)	1	—	1	—	—	5	—	—	—	29.4
132	(2)	0	1	—	—	1	—	—	—	—	80.0
133	(7)	1	3	—	—	4	—	—	—	—	77.8
134	(2)	0	1	—	1	—	—	—	—	—	75.0
135	(1)	1	—	1	—	—	—	—	—	—	00.0
136	(18)	4	7	—	—	11	—	—	—	—	76.6
137	(12)	6	—	9	2	—	1	—	—	—	12.0
138	(4)	1	—	2	1	—	1	—	—	—	22.2
139	(1)	0	—	—	—	—	1	—	—	—	33.3
140	(4)	1	—	1	—	—	3	—	—	—	27.3
141	(3)	0	—	—	—	—	3	—	—	—	33.3
142	(1)	0	—	—	—	—	1	—	—	—	33.3
143	(17)	8	13	—	—	4	—	—	—	—	89.5
144	(10)	4	7	—	—	3	—	—	—	—	87.0
145	(19)	2	4	—	—	15	—	—	—	—	73.1
146	(1)	1	1	—	—	—	—	—	—	—	100.0
147	(7)	6	7	—	—	—	—	—	—	—	100.0
148	(9)	1*	—	—	—	8	—	1	—	—	67.8

TABLE 4
(Continued)

Site	(n) ^a	Males ^b	Genome ^c								%L ^d
			2n			3n		4n			
			LL	JJ	LJ	LLJ	LJJ	LLLJ	LLJJ	LJJJ	
149	(1)	0	—	—	—	1	—	—	—	—	66.7
150	(17)	4	17	—	—	—	—	—	—	—	100.0
151	(9)	4	7	—	—	2	—	—	—	—	90.0
152	(10)	5*	8	—	—	2	—	—	—	—	90.9
153	(1)	0	1	—	—	—	—	—	—	—	100.0
154	(13)	8	13	—	—	—	—	—	—	—	100.0
155	(10)	1	2	—	—	7	—	1	—	—	72.4
156	(13)	5	—	6	3	—	4	—	—	—	23.3
157	(8)	1	—	1	—	—	7	—	—	—	30.4
158	(13)	6	—	7	1	—	5	—	—	—	19.4
159	(9)	1	—	2	1	—	6	—	—	—	29.2
160	(4)	0	—	—	—	—	4	—	—	—	33.3
161	(11)	0	—	—	—	3	7	—	—	1	41.2
162	(16)	5*	5	—	—	10	—	1	—	—	75.0
163	(12)	2	—	3	—	—	9	—	—	—	27.3
164	(12)	5	—	6	—	—	6	—	—	—	20.0
165	(17)	1*	—	—	—	—	17	—	—	—	33.3
166	(15)	2	—	3	—	—	12	—	—	—	28.6
167	(6)	2	—	3	—	—	3	—	—	—	20.0
168	(38)	4	9	—	—	26	—	3	—	—	90.6
169	(7)	1	—	1	1	—	5	—	—	—	31.6
170	(33)	25	—	33	—	—	—	—	—	—	00.0
171	(10)	8	—	10	—	—	—	—	—	—	00.0
172	(9)	5	—	9	—	—	—	—	—	—	00.0
173	(17)	10	—	17	—	—	—	—	—	—	00.0
174	(3)	0	—	—	1	2	—	—	—	—	62.5
175	(21)	15	—	21	—	—	—	—	—	—	00.0
176	(1)	0	—	1	—	—	—	—	—	—	00.0
177	(5)	5	—	5	—	—	—	—	—	—	00.0
178	(1)	0	—	1	—	—	—	—	—	—	00.0
179	(17)	4	—	9	—	—	7	—	—	1	18.6
180	(25)	7	—	25	—	—	—	—	—	—	00.0
181	(16)	10	—	16	—	—	—	—	—	—	00.0
182	(26)	15	—	26	—	—	—	—	—	—	00.0
183	(16)	7	—	16	—	—	—	—	—	—	00.0
184	(2)	2	—	2	—	—	—	—	—	—	00.0
185	(18)	14	—	18	—	—	—	—	—	—	00.0
186	(18)	12	—	18	—	—	—	—	—	—	00.0
187	(12)	4	—	12	—	—	—	—	—	—	00.0
188	(10)	10	—	10	—	—	—	—	—	—	00.0
189	(65)	24	45	—	—	20	—	—	—	—	86.7
190	(4)	4	—	4	—	—	—	—	—	—	00.0
191	(9)	3	—	3	—	—	6	—	—	—	25.0
192	(4)	1	—	1	1	—	1	—	—	1	27.3
193	(3)	0	—	—	—	—	3	—	—	—	33.3
194	(9)	1	—	1	—	—	7	—	—	1	29.6
195	(15)	4	—	6	—	—	9	—	—	—	23.1
196	(3)	1	—	3	—	—	—	—	—	—	00.0
197	(1)	0	—	—	—	1	—	—	—	—	66.7
198	(13)	3	4	—	—	2	4	3	—	—	67.6

TABLE 4
(Continued)

Site	(n) ^a	Males ^b	Genome ^c							%L ^d	
			2n			3n		4n			
			LL	JJ	LJ	LLJ	LJJ	LLLJ	LLJJ		LJJJ
199	(1)	0	1	—	—	—	—	—	—	—	100.0
200	(9)	0	—	—	—	—	9	—	—	—	33.3
201	(7)	0	—	—	7	—	—	—	—	—	50.0
202	(3)	0	—	—	—	—	3	—	—	—	33.3
203	(17)	1	—	1	—	—	11	—	—	5	31.4
204	(20)	14	20	—	—	—	—	—	—	—	100.0
205	(1)	0	—	1	—	—	—	—	—	—	00.0
206	(2)	1	2	—	—	—	—	—	—	—	100.0
207	(12)	0	—	—	—	—	10	—	—	2	31.6
208	(14)	0	—	—	—	—	14	—	—	—	33.3
209	(1)	0	—	—	—	1	—	—	—	—	66.7
210	(5)	3	5	—	—	—	—	—	—	—	100.0
211	(14)	9	10	—	2	2	—	—	—	—	86.7
212	(2)	1	—	2	—	—	—	—	—	—	00.0
213	(1)	0	—	—	—	—	1	—	—	—	33.3
214	(4)	0	1	—	2	1	—	—	—	—	55.6
215	(12)	1	1	—	2	9	—	—	—	—	66.7
216	(1)	0	—	—	—	1	—	—	—	—	66.7
Total	(1377)	453	308	464	86	228	244	26	1	20	
Percentages		32.90	22.37	33.70	6.24	16.56	17.72	1.89	0.07	1.45	
		Percent unisexuals: 43.94				Percent unisexual triploids: 78.02					
		Percent “unisexual males”: 1.32				Percent unisexual tetraploids: 7.77					
		Percent unisexual diploids: 14.21									

^a Sites and individual specimens are provided in appendix 1 and appendix 2.
^b * indicates the finding of a rare “unisexual male” at this site.
^c Electrophoretically identified genotypes of diploid, triploid, and tetraploid salamanders: *Ambystoma laterale* genome (L) and *A. jeffersonianum* genome (J).
^d Percentage of *Ambystoma laterale* genomes (%L) is calculated from all individuals collected at each site.
† New individuals collected from the same sites previously examined by Bogart and Klemens (1997).

presence of the sexual species, we calculated the genomic percentage of *A. laterale* in each site to obtain values ranging from 0% (all *A. jeffersonianum*) to 100% (all *A. laterale*). Under this scheme, a triploid LLJ would be 66.7% (*A. laterale*) and triploid LJJ would be 33.3% (*A. laterale*). If the average percentage of all the individuals from a site was below 50 then *A. jeffersonianum* is assumed to be the sperm donor in that population even though the sample of salamanders may not have included *A. jeffersonianum*. We use the term genomotype (Lowcock, 1994) to describe the template genomic contributions of *A. laterale* (LL) and *A. jeffersonianum* (JJ) in diploid and polyploid unisexual individuals while recognizing that the combination of genomes in unisexuals may be slightly restructured by

intergenomic recombinations and translocations (Bi and Bogart, 2006; Bi et al., 2007).

RESULTS
ELECTROPHORESIS

As in our previous study (Bogart and Klemens, 1997), *A. laterale* could be easily distinguished from *A. jeffersonianum* individuals based on the presence of alternate electrophoretic alleles that were mostly homozygous with a gene frequency (*p*) > 0.90 in both species. The genotypes for *A. laterale* (appendix 2-1) and *A. jeffersonianum* (appendix 2-2) individuals and their allele frequencies for each locus (table 3) demonstrate considerable intraspecific homozygosity for most of

the 11 loci examined, but Sod-1 was the only locus that was alternately fixed ($p = 1.00$) for Sod-1¹⁰⁰ in *A. laterale* and Sod-1³⁷ in *A. jeffersonianum*. Gene frequencies at each of the 11 loci were calculated for the sexual genotypes and unisexual genotypes from all sites (table 3) based on the data for all individuals (appendix 2). The common allele ($p > 0.95$ for *A. jeffersonianum*) was not found in any *A. laterale* specimen for Aat-1⁷⁹, Aat-2⁻¹⁸⁰, Idh-1¹⁴², Mdh-1¹⁷⁶, and Mpi¹²⁰. Based on these diagnostic isozyme alleles and ploidy determination (below), the unisexual specimens were identified as *A. laterale* – *jeffersonianum* (LJ) (appendix 2-3); *A. (2) laterale* – *jeffersonianum* (LLJ) (appendix 2-4); *A. laterale* – (2) *jeffersonianum* (LJJ) (appendix 2-5); *A. (3) laterale* – *jeffersonianum* (LLLJ) (appendix 2-6); *A. laterale* – (3) *jeffersonianum* (LJJJ) (appendix 2-7); and *A. (2) laterale* – (2) *jeffersonianum* (LLJJ) (appendix 2-8). The sites (appendix 1) where the sexual and unisexual individuals were found are included for each individual in appendix 2. A summation of the sexual and unisexual genotypes at each site is provided in table 4. We found more unisexual individuals (44%) than either of the sexual species (23% LL and 34% JJ) and most unisexuals were triploid. Tetraploids (3%) and unisexual males (1%) were rare and were found with unisexual diploid and/or triploid females in scattered populations.

NEW AND RARE ALLELES

Sod-1 was fixed for one allele in *A. laterale* and one in *A. jeffersonianum* but we found additional alleles at the other loci in a few individuals of the two species and the unisexuals. Some rare alleles ($p < 0.05$) were also found in our earlier study in individual *A. laterale* (Aat-1¹¹⁰, Aat-2⁻⁵⁰, Ldh-1⁸⁸, Ldh-2⁵⁵, Mdh-1¹³⁵, Pgi³²⁵, Pgi¹¹⁵, Pgm-1⁹⁷, Pgm-2¹²⁰, Pgm-2⁷⁵) and *A. jeffersonianum* (Aat-1¹⁰⁰, Aat-2⁻¹⁰⁰, Aat-2⁻⁵⁰, Idh-1¹⁰⁰, Ldh-1¹¹⁵, Mdh-1¹⁰⁰, Mpi¹⁴⁰, Mpi¹⁰⁰, Pgi¹⁰⁰, Pgm-1⁸²). In the present study we found most of these same rare alleles and we also found nine new alleles that were not encountered by Bogart and Klemens (1997). A new Ldh-1⁷⁸ allele (D) with a frequency (p) of 0.109 in *A. laterale* (table 3) is not a rare allele. This allele was

found in both *A. laterale* and LLJ unisexual specimens in northern Pennsylvania where it was found in a homozygous condition in *A. laterale* and unisexual LLJ individuals (site 189). Unisexual LLJ from site 148 in western New York were also homozygous for Ldh-1⁷⁸. *Ambystoma laterale* was not collected at that site but is presumed to be the sperm donor (67.8% L, table 4). The other eight new alleles that were found were rare ($p < 0.05$). Aat-1⁸⁶ was found only as a heterozygous allele in one LLJ (site 145) in western New York. Two new Idh-1 alleles were found (Idh-1¹⁶⁰ and Idh-1⁵⁰). The Idh-1¹⁶⁰ allele migrated more anodally on the gel than the Idh-1¹⁴² (A) allele and is designated Q in appendix 2 and table 3. That allele was found in a heterozygote condition in *A. laterale* (sites 113 and 189), *A. jeffersonianum* (sites 118 and 204), and in unisexual LJ and LJJ individuals (site 130). The Idh-1⁵⁰ allele was found in only one *A. laterale* QC heterozygous individual (site 113). A new Ldh-2¹⁶⁰, also a “Q” allele was found in *A. jeffersonianum* heterozygotes (sites 2 and 120) in western New York. This allele was not found in the two individuals from site 2 that were sampled by Bogart and Klemens (1997). Both site 2 and site 120 are in Tompkins County. One LLJ heterozygote from site 208 in eastern New York also possessed the Ldh-2¹⁶⁰ Q allele. A new Mdh-1²⁰⁰ allele was found in *A. jeffersonianum* and an LJJ unisexual from site 158 in Morris County, New Jersey. A new Mpi⁸⁰ allele was found in *A. laterale* from distant localities (sites 108 and 150), *A. jeffersonianum* (site 138), LJ (site 112), and LJJ (site 126). A new Pgi³⁸⁰ Q allele was found in scattered populations that included *A. jeffersonianum* (sites 118, 170, 185) and one LLJ unisexual (site 155). A new Pgm-1¹¹⁵ allele was found in *A. laterale* from western (site 127) and northern New York (sites 150 and 151) as well as in *A. jeffersonianum* from southeastern New York (site 121) and in one unisexual LLJ individual from site 148 in western New York.

HOMOZYGOSITY AND REVERSED GENOTYPES IN UNISEXUALS

The majority of the unisexuals were heterozygous and were identified by the observed

TABLE 5
Unisexual individuals that demonstrated an unexpected homozygous condition for alleles at loci diagnostic for *A. laterale* and *A. jeffersonianum*
Sample sizes in parentheses (n) are the number of individuals of each genomotype that were examined and scored for each locus (from appendix 2) (see footnotes in table 3).

Locus	Mobility	Diploid, triploid, and tetraploid unisexual genomotypes						Total	%
		LJ	LLJ	LJJ	LLLJ	LJJJ	LLJJ		
Aat-2 (n)		(86)	(226)	(242)	(25)	(19)	(1)	(599)	
B	−100						1	(1)	0.17
Idh-1 (n)		(86)	(218)	(241)	(26)	(20)	(1)	(592)	
A	+142	2	3	12		5		(22)	3.72
B	+100	1	16		2			(19)	3.21
Ldh-1 (n)		(85)	(220)	(243)	(26)	(20)	(1)	(595)	
B	+100	2	4		1			(7)	1.18
C	+88	1	1	1				(3)	0.50
D	+78		9					(9)	1.51
Mdh-1 (n)		(84)	(227)	(244)	(26)	(20)	(1)	(602)	
D	+100		2					(2)	0.33
Mpi (n)		(58)	(180)	(186)	(16)	(9)	(1)	(450)	
A	+140		1					(1)	0.22
B	+120	1	1	1	2	1		(6)	1.33
C	+100		11				1	(12)	2.67
Sod-1 (n)		(86)	(226)	(244)	(26)	(20)	(1)	(603)	
B	+100		3		2			(5)	0.83
D	+37	3				1		(4)	0.66

diagnostic alleles at several loci (above). There were, however, a few unisexual individuals that were homozygous for *A. laterale* or *A. jeffersonianum* alleles at some loci. In addition, a few polyploid unisexuals had a genotype that, based on observed electrophoretic dosage, included some reversed patterns. In our earlier investigation (Bogart and Klemens, 1997), we also found homozygous alleles and reversed dosage patterns for some individuals at some loci. Of the six diagnostic loci where homozygous unisexuals were observed, 6.93% of the unisexuals were homozygous for Idh-1 alleles. There were fewer homozygous unisexual individuals at the other loci. The number of unisexual individuals that were homozygous for alleles at one or more of the six diagnostic loci is included in table 5. Again, more unisexual polyploid individuals had a reversed electrophoretic pattern for Idh-1 diagnostic alleles (4.8%), but the frequency of reversals found for any diagnostic allele at

the seven loci involved very few individuals (table 6).

PLOIDY

Data for blood cell measurements for individuals are included in appendix 2 and are summarized in table 7 and fig. 1. Although the ranges for the erythrocyte area measurements were large in all of the genomic classes, a T-test revealed a significant difference ($p < 0.01$) between the ploidy classes when they were grouped as diploid, triploid, or tetraploid. We found a significant difference between measurements of *A. laterale* and *A. jeffersonianum* but no significant difference between the measurements of LJJ triploids and LLLJ tetraploids. These data show that *A. jeffersonianum* has larger blood cells than do *A. laterale* and raise the possibility that measurements of some triploid or tetraploid individuals might not accurately distinguish

TABLE 6
Unisexual individuals that demonstrated a reversed condition for diagnostic alleles
For example, the expected genotype for an LLJ individual for Aat-1 would be Aat-1^{100/100/79} or BBD and a reversed genotype at that locus would be Aat-1^{100/79/79} or BDD which would be the expected genotype for LJJ. A reversal in a tetraploid LLLJ could be Aat-1^{100/100/79/79} or Aat-1^{100/79/79/79}. Sample sizes in parentheses (n) are the number of individuals of each genotype that were examined and scored for each locus (from appendix 2) (see footnotes in table 3).

Locus	Mobility	Diploid, triploid, and tetraploid unisexual genotypes					Total	%
		LLJ	LJJ	LLLJ	LJJJ	LLJJ		
Aat-1 (n)		(227)	(244)	(26)	(20)	(1)	(518)	
B	+100		1		0	0		0.19
D	+79	5		0		0	5	0.96
Aat-2 (n)		(223)	(242)	(25)	(20)	(1)	(505)	
A	-180	1		0		0	1	0.20
B	-100		6		0	0	6	1.19
Idh-1 (n)		(218)	(241)	(26)	(20)	(1)	(499)	
A	+142	4		1		0	5	1.00
B	+100		22		0	0	22	4.41
Ldh-1 (n)		(220)	(243)	(26)	(20)	(1)	(504)	
B	+100		12		2	0	14	2.58
C	+88	7		1		0	8	1.39
Mdh-1 (n)		(227)	(244)	(26)	(20)	(1)	(512)	
B	+176	15		0		0	15	2.93
D	+100		1		0	0	1	0.20
Mpi (n)		(180)	(186)	(16)	(9)	(1)	(387)	
B	+120	2		0		0	2	0.52
C	+100		7		0	0	7	1.81
Sod-1 (n)		(226)	(235)	(26)	(20)	(1)	(511)	
B	+100	0		0		0	0	0.00
D	+37		0		0	0	0	0.00

these ploidy classes. We have more confidence in the ploidy determinations based on flow cytometric data, but those data were not quantified as absolute picograms of DNA. Blood cells from individuals were compared with a diploid *A. jeffersonianum* standard diploid peak (Ramsden et al., 2006) to determine ploidy. Ploidy determinations for such individuals are signified by FCM in appendix 2.

CONNECTICUT AND MASSACHUSETTS

Unisexuals were found in 14 of the 19 sites sampled in Connecticut and Massachusetts in the present study. We found one site (196) in Hartford County, Connecticut, that only had

A. jeffersonianum (n = 3), but the sample size is too low to confirm a pure *A. jeffersonianum* population. We found only one *A. laterale* at another Hartford County site (199). Windham County sites (60: n = 2; 204: n = 20) in Connecticut as well as a Bristol and Plymouth County site (66: n = 2) in Massachusetts are probably pure *A. laterale* sites. Only *A. laterale* was found in our earlier study (Bogart and Klemens, 1997) at site 60 (n = 17) and site 66 (n = 4). In our present study, only *A. laterale* and LLJ individuals were collected in Massachusetts. Many more sites in these states were sampled by Bogart and Klemens (1997), and the distribution provided in figure 6 includes our earlier findings. Both *A. laterale* as well as *A. jeffersonianum* and

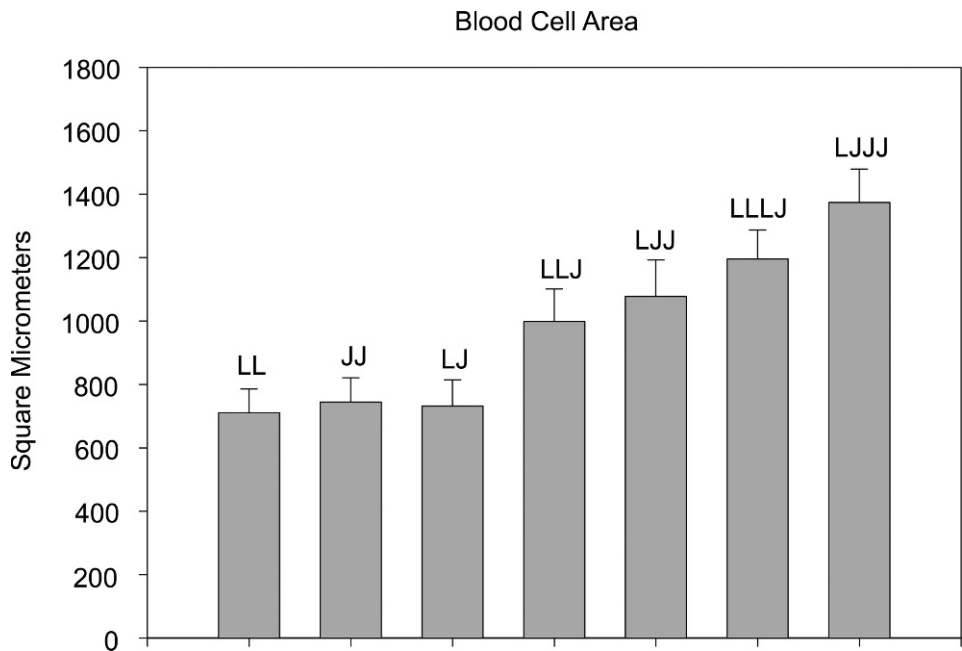


Fig. 1. Graph of mean (\pm SD) erythrocyte area for diploid *Ambystoma jeffersonianum* (JJ), diploid *A. laterale* (LL), diploid unisexuals (LJ), triploid unisexuals (LJJ, LLJ), and tetraploid unisexuals (LLLJ, LJJJ) from the data in table 7.

TABLE 7
Summation statistics for blood cell measurements for *A. laterale* (LL), *A. jeffersonianum* (JJ), and unisexual individuals prior to the use of flow cytometric methods (FCM) for ploidy determination
Average erythrocyte values for the individuals are in appendix 2.

Erythrocyte Area Measurements (μm ²)			
Genome ^a	(n)	Mean ± SD	Range
Diploids			
LL	(205)	710.80 ± 74.89	515 — 949
JJ	(392)	744.54 ± 76.26	387 — 1063
LJ	(69)	732.16 ± 82.85	651 — 978
Triploids			
LLJ	(177)	998.67 ± 102.73	670 — 1252
LJJ	(162)	1077.89 ± 114.75	731 — 1503
Tetraploids			
LLLJ	(14)	1195.28 ± 91.94	1016 — 1317
LJJJ	(7)	1374.00 ± 105.26	1203 — 1536

^a Genomes include haploid complements of *Ambystoma laterale* (L) and *A. jeffersonianum* (J).

their unisexual associates are widespread in these states.

NEW JERSEY

Ambystoma laterale, *A. jeffersonianum*, and unisexuals have previously been reported to occur in New Jersey (Uzzell, 1964; Anderson and Giacosis, 1967). Unisexuals were found in 15 of the 17 New Jersey sites that we sampled (appendix A). The only site where unisexuals were not collected was an *A. laterale* site in Morris County (site 154; n = 13) in northern New Jersey. Even though more *A. jeffersonianum* individuals (n = 31) were collected from more sites in New Jersey than were those of *A. laterale* (n = 28) (table 4), no site was found for *A. jeffersonianum* that did not also have unisexual individuals (fig. 4).

NEW YORK

Unisexuals were found in most (43 of 57) New York sites (appendix 1) (fig. 5). The northern New York sites in St. Lawrence County (sites 149 to 152) were dominated by



Fig. 2. *Ambystoma laterale* and unisexuals associated with *A. laterale* found in Pennsylvania. *Ambystoma laterale* (AMNH 165901 from site 189) (top), diploid unisexual LJ (AMNH 169928 from site 174) (middle), and triploid unisexual LLJ (AMNH 166022 from site 189) (bottom).

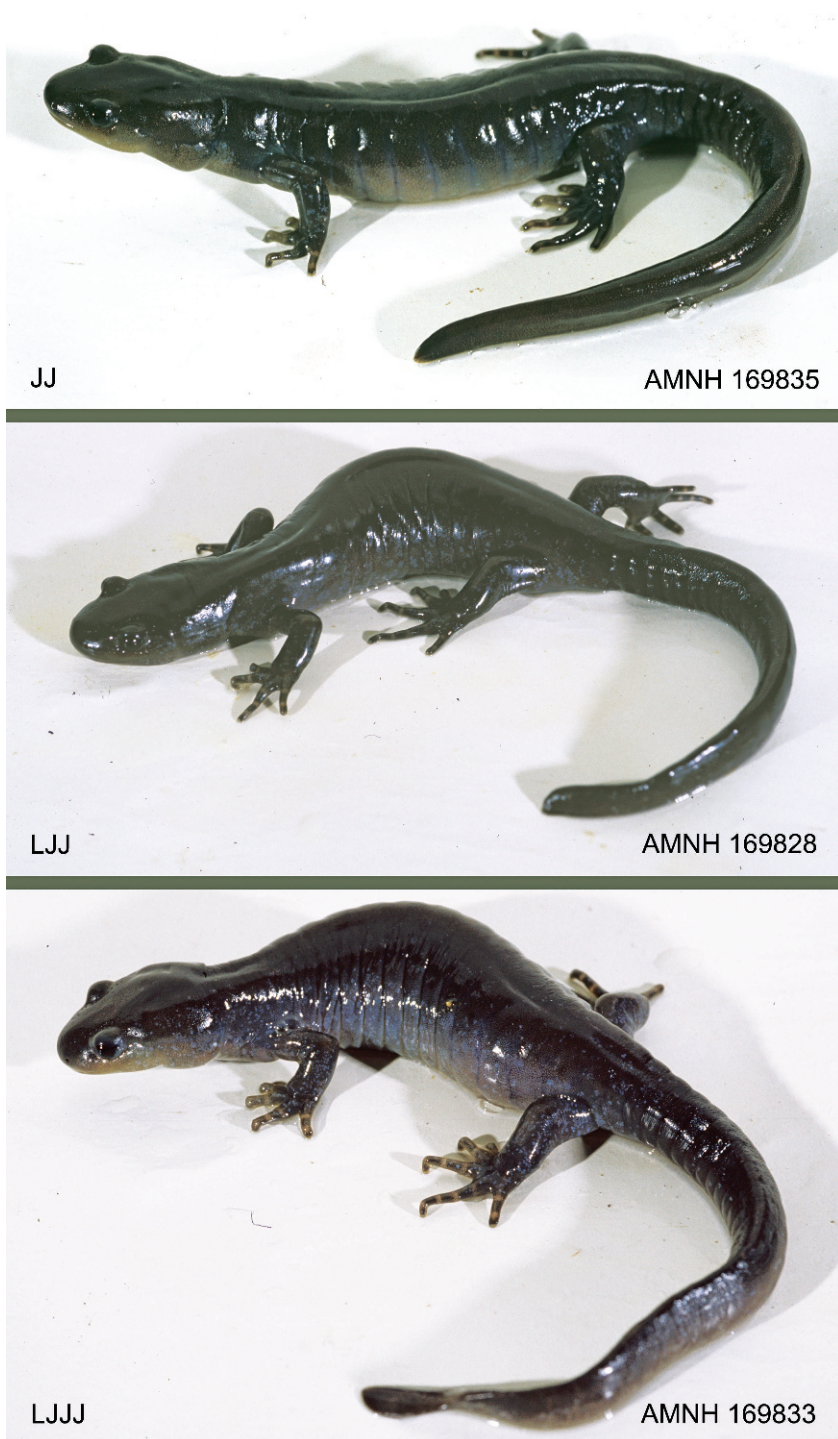


Fig. 3. *Ambystoma jeffersonianum* and unisexuals associated with *A. jeffersonianum* found in Pennsylvania. *Ambystoma jeffersonianum* (AMNH 169835) (top), triploid unisexual (LJJ) (AMNH 169828) (middle), and tetraploid LJJJ unisexual (AMNH 169833) (bottom). All are from site 179.

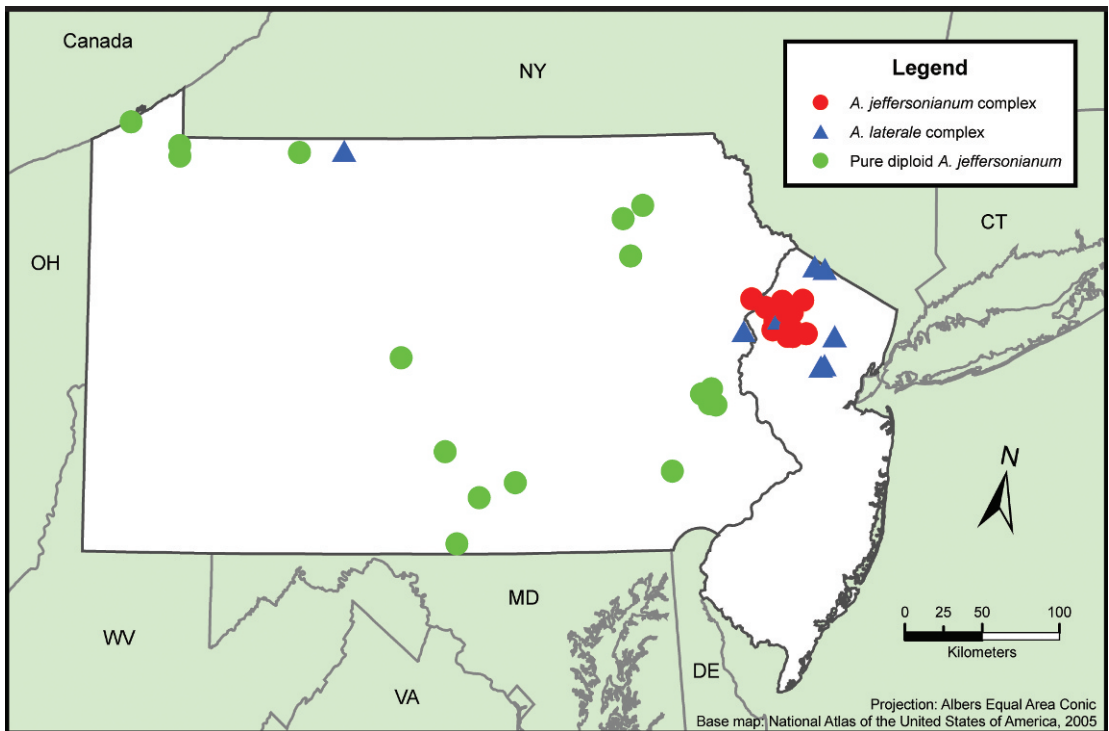


Fig. 4. Distribution of *Ambystoma jeffersonianum*, *Ambystoma laterale*, and unisexuals in New Jersey and Pennsylvania.

A. laterale, and only *A. laterale* ($n = 17$) was found at site 150. Unisexual LLJ were found with *A. laterale* in sites 151 (LL: $n = 7$; LLJ: $n = 2$) and 152 (LL: $n = 8$; LLJ: $n = 2$). A single LLJ individual was collected at site 149. Other sites where we found only *A. laterale* were in western New York counties of Erie (site 146: $n = 1$) and Niagara (site 147: $n = 7$); in the northeastern county of Essex (site 153: $n = 1$); and in the southeastern counties of Dutchess (site 206: $n = 2$) and Putnam (site 210: $n = 5$), but with such small sample sizes, these sites could also contain unisexual individuals. *Ambystoma jeffersonianum* were found in populations throughout New York (fig. 5). Populations containing only *A. jeffersonianum* were Tompkins County (site 2: $n = 38$; site 120: $n = 14$), Sullivan County (site 121: $n = 20$), and Otsego County (site 118: $n = 16$) in east-central New York. The single *A. jeffersonianum* specimens from site 116 in Ulster County is probably not a pure *A. jeffersonianum* site because site 116 is in the same drainage basin and is close to site 117 where

LJJ and LJJJ individuals were found. Other New York sites where only *A. jeffersonianum* were found were represented by only one or two individuals: Cattaraugus County in western New York (site 135: $n = 1$) and Columbia County (site 212: $n = 2$), as well as Westchester County (site 205: $n = 1$) in southeastern New York.

PENNSYLVANIA AND VIRGINIA

Ambystoma laterale and unisexuals have not been reported to occur in Pennsylvania. As expected, most of the Pennsylvania sites contained only *A. jeffersonianum*, but unisexual LJJ ($n = 7$) and LJJJ ($n = 1$) were found with *A. jeffersonianum* ($n = 9$) at site 179 in Monroe County in eastern Pennsylvania. *Ambystoma jeffersonianum*, LJJ, and LJJJ from site 179 are shown in figure 3. We also found one LJ and three LLJ unisexual individuals at site 174 in Northampton County, which is also in eastern Pennsylvania. Individual *A. laterale* were not found at site

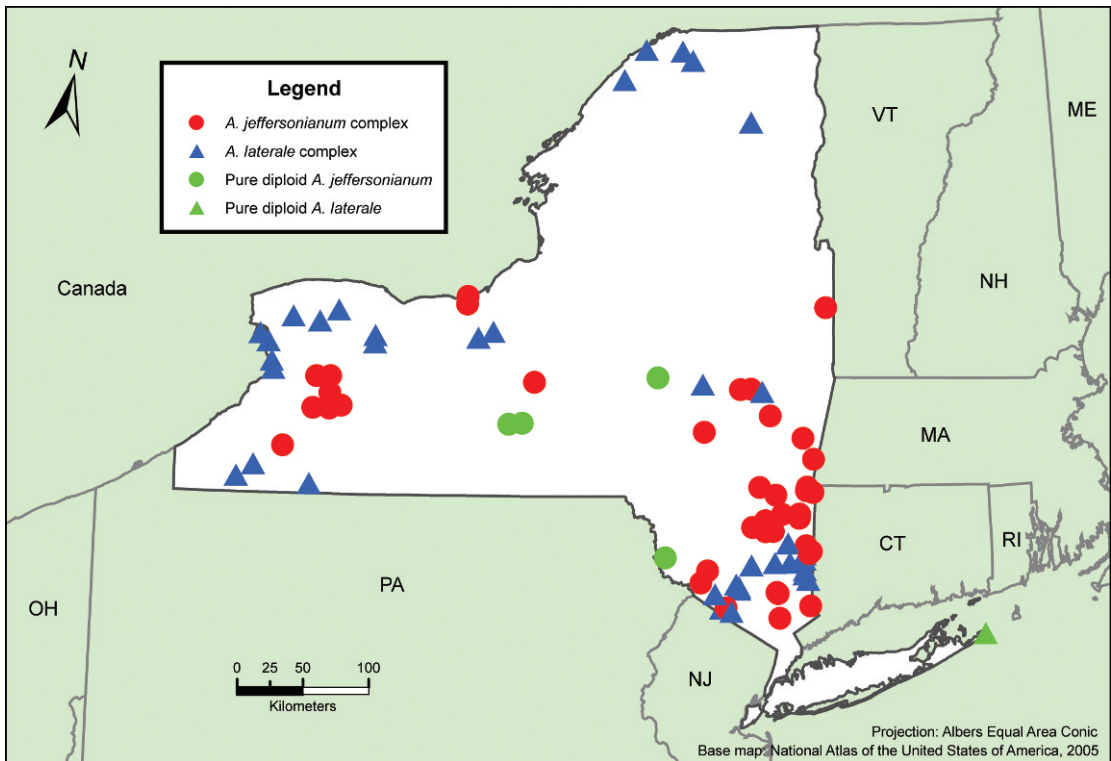


Fig. 5. Distribution of *Ambystoma jeffersonianum*, *Ambystoma laterale*, and unisexuals in New York.

174 but is assumed to be the sperm donor for these unisexuals (62.5% L, table 4). *Ambystoma laterale* (n = 45) was found in McKean County (site 189), in north-central Pennsylvania, with LLJ (n = 20) unisexuals. *Ambystoma laterale*, LLJ, and LLJ individuals from these sites are provided in figure 2. The distribution of sexual and unisexual individuals that we found in Pennsylvania is plotted in fig. 4. Only four individuals were sampled from one site (site 190) in Virginia. They were all *A. jeffersonianum*.

DISCUSSION

We found a lower ratio of unisexual to sexual individuals than were found by Bogart and Klemens (1997) in populations from New England and mostly eastern New York. We can account for the discrepancy because, in the present study, we sampled large numbers of *A. jeffersonianum* from populations in Pennsylvania where unisexuals probably do

not exist and we sampled some more northern populations in New York where unisexuals were found to be at a lower frequency than *A. laterale*. But, in keeping with our previous study, in most populations where unisexuals coexist with sexual individuals, the unisexuals are still more numerous. During the course of this study, we visited some of the same sites in different years to increase our sampling of populations to find rare genotypes, which can confirm the absence or presence of sexual and unisexual individuals. However, this was not always possible, especially if the salamanders were rare or difficult to collect and many sites are represented by one or two individuals. Because the unisexuals require a sperm donor to successfully reproduce, if a sexual individual was not encountered, we predicted the presence of *A. laterale* or *A. jeffersonianum* based on the genotypes of the unisexuals. It was not possible to predict the absence or the presence of unisexuals. The possibility exists that a population might change over time or

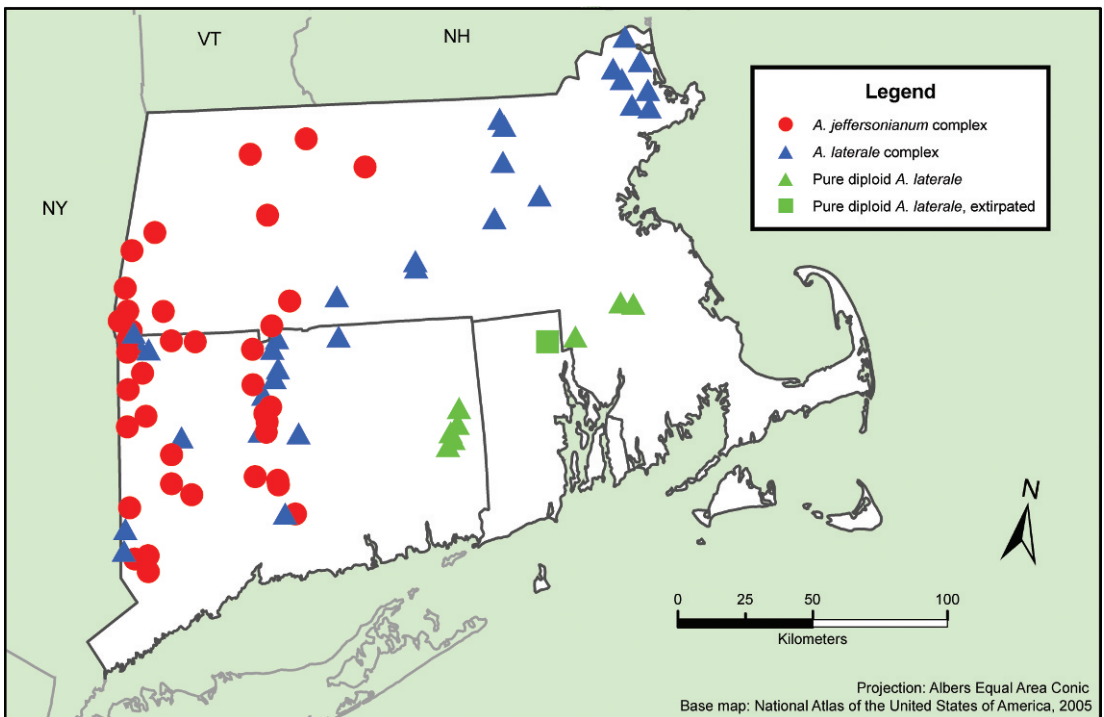


Fig. 6. Distribution of *Ambystoma jeffersonianum*, *Ambystoma laterale*, and unisexuals in Connecticut, Massachusetts, and Rhode Island.

that our sampling was biased. Eight of the 118 sites (marked † in table 4) sampled in the present study were also sampled for our earlier study (Bogart and Klemens, 1997). The species and genotypes of the unisexuals were similar for most of the resampled populations. Increasing the sample size from three (Bogart and Klemens, 1997) to 38 individual *A. jeffersonianum* at site 2 in western New York supported our earlier contention that unisexuals are absent at that site. We did find *A. jeffersonianum* in Hartford County at Granby (site 195) and King Phillip Mountain near Simsbury (site 196), but we also found *A. laterale* at East Granby (site 197) and Farmington (site 198) near Rattlesnake Mountain. The Burnt Hill site (site 199), also near Farmington, yielded a single LJJ. A site near Wethersfield (site 201) was unusual as we found only seven diploid LJ unisexuals and, therefore, we can not predict a putative sperm donor for that site. Evidently, both sexual species are present and in close proximity in this region of Hartford County.

NO APPARENT SYNTOPIC ASSOCIATION OF THE SEXUAL SPECIES

Ambystoma laterale and *A. jeffersonianum* were not found together in any of the 118 sites and there were only four sites where both LLJ and LJJ were found (sites 108 and 113 in New York, site 161 in New Jersey, and site 197 in Connecticut) (appendix 1; table 4). Both New York sites were in Orange County. At site 108 we found LJJ ($n = 6$), LLJ ($n = 17$), LLLJ ($n = 3$), and LJJJ ($n = 3$) unisexual individuals. *Ambystoma laterale* ($n = 14$) was the only sperm donor collected at that site, but the LJJ triploids and the LJJJ tetraploids indicate the sympatric presence of *A. jeffersonianum*. Based on egg mass data (Bi and Bogart, 2006; Bi et al., 2007; Bogart et al., 2007) tetraploids are often found among the triploid offspring from individual triploid unisexuals through ploidy elevation that may be mediated by elevated temperature (Bogart et al., 1989). The additional J genome in the LJJJ tetraploids would be an indication that *A.*

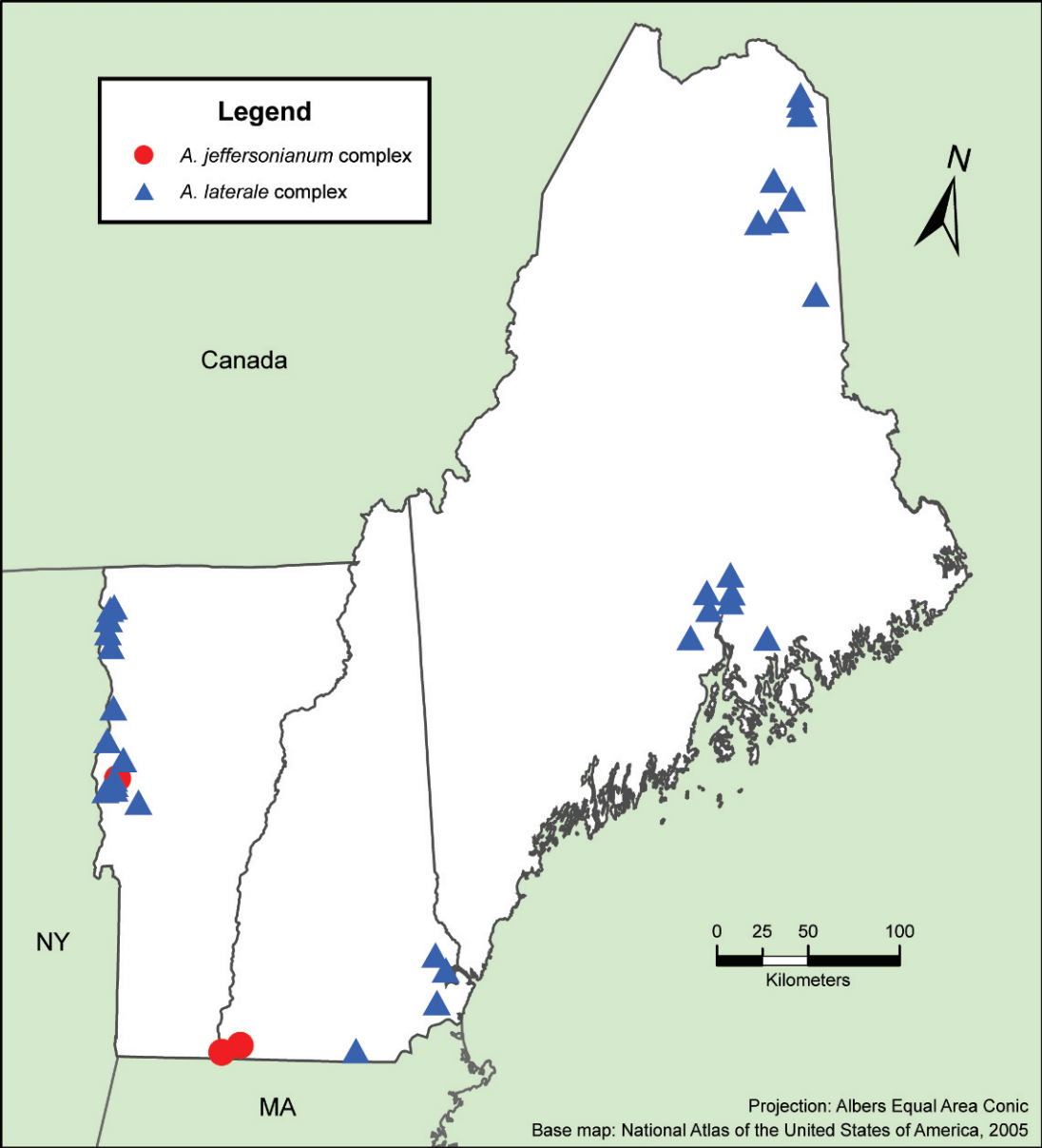


Fig. 7. Distribution of *Ambystoma jeffersonianum*, *Ambystoma laterale*, and unisexuals in Maine, New Hampshire, and Vermont.

jeffersonianum was the sperm donor if the LJJJ unisexual individuals were derived from a triploid LJJ female, but the tetraploids could have been gynogenetic offspring from a tetraploid LJJJ female. *Ambystoma laterale* and associated LLJ and LLLJ unisexuals were found in other Orange County sites 107, 110,

111, and 113. *Ambystoma jeffersonianum*, LJ, and LJJ were found in Orange County (site 114). If *A. jeffersonianum* does occur at site 108, it must be less common than *A. laterale* and that population might demonstrate the phenomenon of one sexual species displacing the other over time. Making a case for the

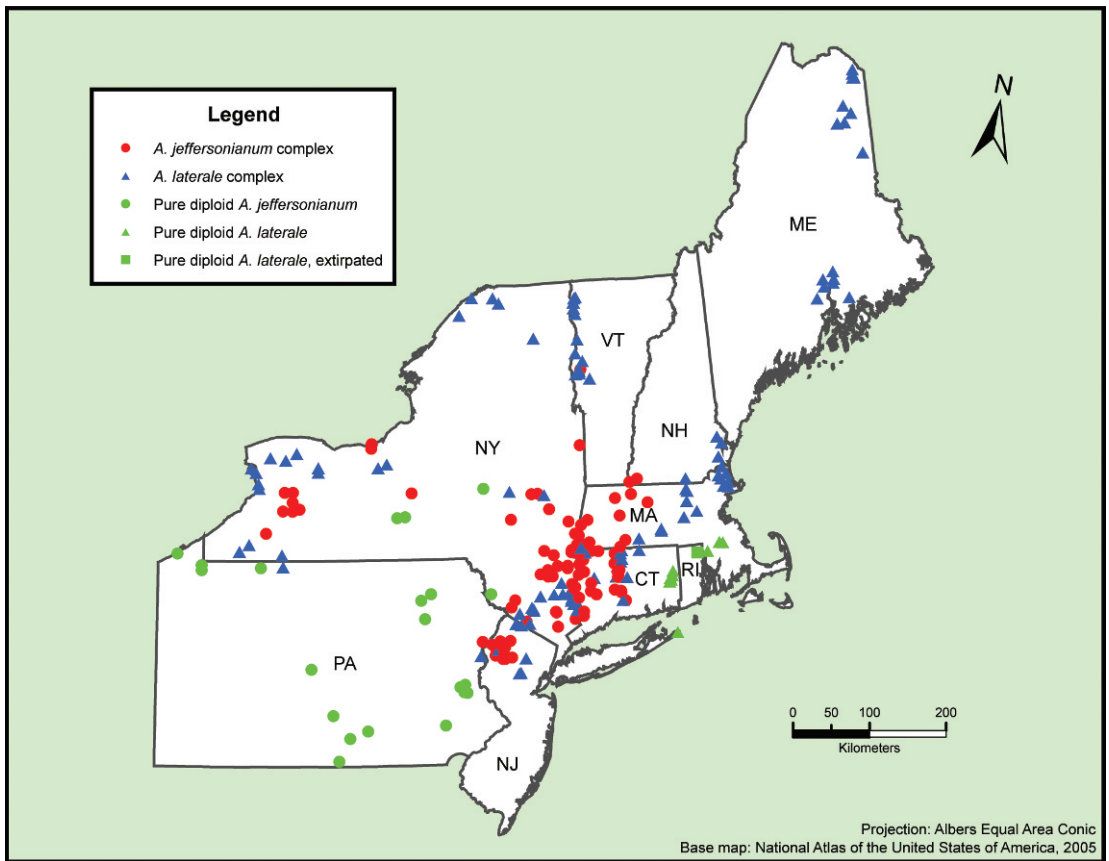


Fig. 8. Distribution of *Ambystoma jeffersonianum*, *Ambystoma laterale*, and unisexuals in northeastern U.S.

presence of *A. jeffersonianum* in the other Orange County site that is dominated by *A. laterale* (site 113) is more tenuous because, of the 43 specimens collected at that site, there was only one LJJ individual and no LJJJ tetraploids. Dispersal of LJJ unisexuals into an *A. laterale*/unisexual site would be a possible alternate explanation.

Neither sexual species was collected at site 161 in Warren County, New Jersey, where LLJ ($n = 3$) and LJJ ($n = 7$) were found. The tetraploid LJJJ that was also collected at site 161 provides similar evidence that *A. jeffersonianum* is most likely the sperm donor. In that same county, both *A. laterale* ($n = 5$) (site 162) and *A. jeffersonianum* (site 159: $n = 2$; site 163: $n = 3$) were also collected. Based on our collection, the Hartford, Connecticut, site (198) is an *A. laterale*/unisexual site because *A.*

laterale ($n = 4$) and three LJJJ tetraploids were collected, but the three LJJ unisexuals that were also collected could indicate that this site also has *A. jeffersonianum* as have been found in other Hartford County sites (195 and 196).

From the relatively few examples of the sympatric occurrence of LLJ and LJJ, it is evident that all such instances exist only where *A. laterale* and *A. jeffersonianum* are parapatric and possibly sympatric over time and space. In a southern Ontario population where *A. laterale* and *A. jeffersonianum* were found to be breeding in the same pond, LJJ and LLJ unisexuals shared microsatellite alleles (Bogart et al., 2007). In that study, *A. laterale* was found to be an acceptable sperm donor for LJJ unisexual females because LLJJ larvae were encountered in the same egg mass with LJJ larvae. Through genome replacement, the

switch from LJJ to LLJ or vice versa is probably a rapid and widespread phenomenon within the unisexual complex (Bogart, 2003) and would explain our observations in the few populations where this process is probably occurring.

DIPLOID AND TETRAPLOID UNISEXUALS

Most unisexuals in this and previous studies (Uzzell, 1964; Bogart et al., 1987; Bogart and Klemens, 1997) are triploid. Tetraploids can be produced in the laboratory (Bogart et al., 1989) and are found to be derived in nature from triploid individuals (Bogart et al., 2007) through the process of ploidy elevation where a sperm nucleus is incorporated in an unreduced, triploid egg. We found tetraploids in scattered populations (table 4) and triploids were always found in the same populations. Diploid unisexuals are much more difficult to explain. They, too, are scattered in various populations (table 4) and, in a few populations, they were more numerous than the triploids (e.g., sites 123, 125, 137, 214) or were the only unisexual genomotype found (sites 134, 201). They occur in sites with either *A. laterale* or *A. jeffersonianum*. Diploid LJ and triploid LLJ or LJJ unisexuals share the same microsatellite alleles when they are sympatric and diploid LJ unisexuals can produce triploids through ploidy elevation events (Bogart et al., 2007). Ploidy reductional events probably occur from triploid to diploid unisexuals, but empirical evidence is lacking. Two very interesting sites (sites 123 and 125) in Schoharie County, New York, have LJ and LLJ unisexuals as well as *Ambystoma jeffersonianum*. The other Schoharie site (124), which also has *A. jeffersonianum* and LJ unisexuals, has the expected LJJ unisexual genomotype. We did not find any *A. laterale* in Schoharie County. Nor did we find *A. laterale* in Albany (site 126) or Otsego (site 118) counties that are, respectively, north and south of Schoharie County. However, in our 1997 paper we found *A. laterale* and its associated hybrids in Albany County (site 3). Only *A. jeffersonianum* ($n = 16$) was found at site 118. We found *A. jeffersonianum* ($n = 4$), as well as LJ ($n = 5$), LJJ ($n = 18$), and LJJJ ($n = 2$) unisexuals at site 126.

"UNISEXUAL" MALES

We found a very low frequency of "unisexual" males in this investigation and in our earlier study (Bogart and Klemens, 1997). They were diploid LJ (sites 114, 122), triploid LLJ (sites 108, 112, 152, 162), triploid LJJ (Site 165), and tetraploid LLLJ (site 148). Such males are probably sterile based on the limited cytological evidence of meiotic chromosomes (Bogart, 2003). It is possible that an answer to the rare finding of "unisexual" males may be found among the chromosome translocations and recombinations (Bi and Bogart, 2006; Bi et al., 2007) that may involve sex-determining genes, but sex-determining genes have not been mapped in unisexuals to any chromosome region or, indeed, to any chromosome. The possibility exists that these rare males, even if they are sterile, could stimulate gynogenetic development of unisexual eggs. This might be important if other male sexual sperm donors are not available in a breeding pond. Obviously, natural selection should favor the production of female unisexuals.

HOMOZYGOSITY AND REVERSALS OF DIAGNOSTIC ALLELES IN UNISEXUAL INDIVIDUALS

Identification of unisexual genomotypes is based on observations of allozymes that are diagnostic for *A. laterale* and *A. jeffersonianum* and demonstrate the appropriate dosage in diploids and polyploids. Homozygous allozyme patterns at these loci identify the sexual species and a reversed dosage pattern of allozymes at a locus would misidentify a LLJ as an LJJ or vice versa. Misidentification and ambiguity is alleviated by using several loci that possess diagnostic allozymes, so it is possible to recognize that reversals can and do occur, but if an individual had a reversed pattern for several loci it could be misidentified. If unisexuals were identical, genetic clones, as has been previously proposed (Uzzell, 1964; Macgregor and Uzzell, 1964; Uzzell and Goldblatt, 1967), a unisexual lineage could not revert from a homozygous to a heterozygous condition and reversed patterns, once attained, would be fixed (Asher and Nace, 1971). Fluorescent genomic in situ hybridization (GISH) of unisexual

chromosomes (Bi and Bogart, 2006; Bi et al., 2007) and microsatellite analyses (Bogart et al., 2007) clearly show that unisexuals are not clones and, based on chromosomal translocations and recombinations, may be expected to demonstrate gene restructuring. Thus, we may be surprised that observed homozygous and reversed-allozyme alleles are not more common and widespread than the few instances that we found in the present study (tables 5 and 6) and in our earlier study (Bogart and Klemens, 1997). We suspect that natural selection eliminates many such mutational events and helps to explain the embryonic mortality that is ubiquitous in unisexual populations. Unisexual kleptogens swap genomes with sexual sperm donors (Bi et al., 2008). This process may be essential for unisexuals to recover from detrimental genetic restructuring and consequentially, maintain a fixed heterozygous genotype that is most commonly found in unisexual individuals and likely has a selective advantage.

RARE ALLELES

Loci chosen to identify the sexual and unisexual individuals were loci that were mostly monomorphic for alternate alleles in *A. jeffersonianum* and *A. laterale*, but these same loci are not considered to be very conservative in other vertebrates. When sampling individuals from many populations over a large range we should expect to find polymorphism and population specific "private" alleles. The very low frequency of some rare alleles may be attributed to mutational events and especially if a single rare allele is found in only one individual. Finding rare alleles among sexual and unisexual individuals in the same populations adds credence to kleptogenesis.

Some rare alleles that were found in both *A. jeffersonianum* and *A. laterale* such as Aat-2⁻⁵⁰, Mdh-1²⁰⁰ or Mpi⁸⁰ (table 3) may be alleles that have been passed on from a common ancestor and are maintained at a low frequency in both species. A rare allele demonstrates that a diploid-triploid unisexual relationship exists in site 130. The only unisexuals that were found to have Idh-1¹⁶⁰ were LJ at site 130 and LJJ from sites 130 and

131. That allele was not found in *A. jeffersonianum* from those sites but was found in individual *A. jeffersonianum* in sites 118 and 204 and in *A. laterale* from sites 113 and 189. Ldh-1⁷⁸ was not found by Bogart and Klemens (1997). We found this allele in *A. laterale* populations (sites 127, 134, 136, 143, 145) in western New York and in site 189 in north-central Pennsylvania. The unisexual LLJ individuals from the Pennsylvania site that also had this allele were homozygous for Ldh-1⁷⁸. LLJ homozygotes for this allele were also found in site 148 close to Lake Erie in New York. The relatively high frequency of Ldh-1⁷⁸ in western New York *A. laterale* individuals is a clear indication that *A. laterale* genomes that contain this allele have been used to replace L genomes in sympatric unisexuals. The homozygous condition of Ldh-1⁷⁸ in the unisexuals demonstrates that the diagnostic *A. jeffersonianum* Ldh-1⁸⁸ allele was probably lost through genome translocations or recombination or is no longer functional in the homozygous unisexuals.

SUMMARY AND CONCLUSIONS

Identification of *Ambystoma jeffersonianum* and *A. laterale* and their breeding ponds in the northeastern United States in this, and in our earlier, study should benefit conservation efforts to preserve and maintain these species. Populations that we examined as many as 20 years ago may no longer exist. The interaction of unisexuals with the sexual species is a fascinating evolutionary phenomenon that is probably unique. Unisexual kleptogens take genomes from either sexual species and are usually more numerous in sympatric association with a sexual species. Unisexuals were not found in most of the *A. jeffersonianum* populations in Pennsylvania and are less numerous than *A. laterale* in northern New York populations. Perhaps the unisexuals are recent immigrants in those New York populations and have not yet dispersed throughout Pennsylvania. The fact that no unisexuals were found with *A. laterale* in Long Island (New York), eastern Connecticut, and southeastern Massachusetts (Bogart and Klemens, 1997) could also be the result of isolation of those populations prior to unisexual dispersion.

Although no new samples were obtained from Vermont, New Hampshire, and Maine, we have included the ranges of known sexual and unisexual individuals from those states in figure 7 from our earlier investigation (Bogart and Klemens, 1997). Figure 8 includes the known ranges for these salamanders in the northeastern United States from the present and previous study.

In addition to *A. laterale* and *A. jeffersonianum*, unisexuals use other species (*A. texanum*, *A. tigrinum*) as sperm donors in Ohio, Illinois, Indiana, and Michigan (Kraus, 1995; Morris, 1985; Kraus and Petranksa, 1989; Selander, 1994; Bogart, 2003). Distinguishing sexual and unisexual individuals in those states is complicated because *A. texanum* and *A. laterale* have the same allozymes for some of the diagnostic loci (Bogart et al., 1987). It is likely that isozyme electrophoresis will be replaced as a technique to identify these salamander species and their associated unisexuals. Sequences of mitochondrial DNA show all of the sexual species to be distinctive and distinctly different from the unisexuals (Hedges et al., 1992; Bogart, 2003; Bogart et al., 2007). Microsatellite DNA alleles can identify *A. jeffersonianum*, *A. laterale* and the unisexuals that use those species as sperm donors (Julian et al., 2003; Ramsden et al., 2006; Bogart et al., 2007). Because these DNA techniques do not require sacrificing individuals, there are obvious advantages when attempting to identify rare, threatened, or endangered salamanders. Identifying the species and unisexuals is a prerequisite to protecting the species and their habitat. Subsequent temporal monitoring will be necessary to understand the interactions of the species and the unisexual associates.

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REFERENCES

- Anderson, J.D., and R.V. Giacosis. 1967. *Ambystoma laterale* in New Jersey. *Herpetologica* 23: 109–111.
- Asher, J.H., and G.W. Nace. 1971. The genetic structure and evolutionary fate of parthenogenetic amphibian populations as determined by Markovian analysis. *American Zoologist* 11: 381–398.
- Austin, N.E., and J.P. Bogart. 1982. Erythrocyte area and ploidy determination in the salamanders of the *Ambystoma jeffersonianum* complex. *Copeia* 1982: 485–488.
- Bi, K., and J.P. Bogart. 2006. Identification of intergenomic recombinations in unisexual salamanders of the genus *Ambystoma* by genomic *in situ* hybridization. *Cytogenetic and Genome Research* 112: 307–312.
- Bi, K., J.P. Bogart, and J. Fu. 2007. Intergenomic translocations in unisexual salamanders of the genus *Ambystoma* (Amphibia, Caudata). *Cytogenetic and Genome Research* 116: 289–297.
- Bi, K., J.P. Bogart, and J. Fu. 2008. The prevalence of genome replacement in unisexual salamanders of the genus *Ambystoma* (Amphibia, Caudata) revealed by nuclear gene genealogy.

- BMC Evolutionary Biology 8: 158. (doi:10.1186/1471-2148-8-158).
- Bogart, J.P. 1982. Ploidy and genetic diversity in Ontario salamanders of the *Ambystoma jeffersonianum* complex revealed through and electrophoretic examination of larvae. Canadian Journal of Zoology 60: 848–855.
- Bogart, J.P. 1989. A mechanism for interspecific gene exchange via all-female salamander hybrids. In R.M. Dawley and J.P. Bogart (editors), Evolution and ecology of unisexual vertebrates. New York State Museum Bulletin 466: 170–179.
- Bogart, J.P. 2003. Genetics and systematics of hybrid species. In D.M. Sever (editor), Reproductive biology and phylogeny of Urodela, vol. 1, 109–134. Enfield, NH: M/s Science.
- Bogart, J.P., K. Bi, J. Fu, D. Noble, and J. Niedzwiecki. 2007. Unisexual salamanders (genus *Ambystoma*) present a new reproductive mode for eukaryotes. Genome 50: 119–136.
- Bogart, J.P., R.P. Elinson, and L.E. Licht. 1989. Temperature and sperm incorporation in polyploid salamanders. Science 246: 1032–1034.
- Bogart, J.P., and M.W. Klemens. 1997. Hybrids and genetic interactions of mole salamanders (*Ambystoma jeffersonianum* and *A. laterale*) (Amphibia: Caudata) in New York and New England. American Museum Novitates 3218: 1–78.
- Bogart, J.P., and L.E. Licht. 1986. Reproduction and the origin of polyploids in hybrid salamanders of the genus *Ambystoma*. Canadian Journal of Genetics and Cytology 28: 605–617.
- Bogart, J.P., L.E. Licht, M.J. Oldham, and S.J. Darbyshire. 1985. Electrophoretic identification of *Ambystoma laterale* and *Ambystoma texanum* as well as their diploid and triploid interspecific hybrids (Amphibia: Caudata) on Pelee Island, Ontario. Canadian Journal of Zoology 63: 340–347.
- Bogart, J.P., L.A. Lowcock, C.W. Zeyl, and B.K. Mable. 1987. Genome constitution and reproductive biology of the *Ambystoma* hybrid salamanders on Kelleys Island in Lake Erie. Canadian Journal of Zoology 65: 2188–2201.
- Clayton, J.W., and D.N. Tretiak. 1972. Amine-citrate buffers for pH control in starch gel electrophoresis. Journal of the Fisheries Research Board of Canada 29: 1169–1172.
- Danzmann, R.G., and J.P. Bogart. 1982. Gene dosage effects on MDH isozyme expression in diploid, triploid, and tetraploid treefrogs of the genus *Hyla*. Journal of Heredity 73: 277–280.
- Downs, F.L. 1978. Unisexual *Ambystoma* from the Bass Islands of Lake Erie. Occasional Papers of the Museum of Zoology University of Michigan 685: 1–36.
- Drowne, F.P. 1905. The reptiles and batrachians of Rhode Island. Roger Williams Park Museum Monograph 15: 1–24.
- Hedges, S.B., J.P. Bogart, and L.R. Maxson. 1992. Ancestry of unisexual salamanders. Nature 356: 708–710.
- International Union of Biochemistry Nomenclature Committee (IUBC). 1984. Enzyme nomenclature. New York: Academic Press.
- Julian, S.E., T.L. King, and W.K. Savage. 2003. Novel Jefferson salamander, *Ambystoma jeffersonianum*, microsatellite DNA markers detect population structure and hybrid complexes. Molecular Ecology Notes 3: 95–97.
- Kezer, J., and S.K. Sessions. 1979. Chromosome variation in the plethodontid salamander *Aneides ferreus*. Chromosoma 71: 65–80.
- Klemens, M.W. 1978. Variation and distribution of the turtle, *Chrysemys picta*, (Schneider), in Connecticut. MS thesis, University of Connecticut, Storrs, 60 pp.
- Klemens, M.W. 1993. The amphibians and reptiles of Connecticut and adjacent regions. State Geological and Natural History Survey of Connecticut Bulletin 112: 1–318 + 32 plates.
- Klemens, M.W. 2000. Amphibians and reptiles in Connecticut: a checklist with notes on conservation status, identification, and distribution. DEP Bulletin 32: 1–96, Hartford, CT: Connecticut Department of Environmental Protection, Environmental and Geographic Information Center.
- Kraus, F. 1995. The conservation of unisexual vertebrate populations. Conservation Biology 9: 956–959.
- Kraus, F., and J.W. Petranks. 1989. A new sibling species of *Ambystoma* from the Ohio River drainage. Copeia 1989: 94–110.
- Lowcock, L.A. 1994. Biotype, genototype, and genotype: variable effects of polyploidy and hybridity on ecological partitioning in a bisexual-unisexual community of salamanders. Canadian Journal of Zoology 72: 104–117.
- Lowcock, L.A., and J.P. Bogart. 1989. Electrophoretic evidence for the multiple origins of triploidy in the *Ambystoma laterale-jeffersonianum* complex. Canadian Journal of Zoology 67: 350–356.
- Lowcock, L.A., L.E. Licht, and J.P. Bogart. 1987. Nomenclature in hybrid complexes of *Ambystoma*: no case for the erection of hybrid “species”. Systematic Zoology 36: 328–336.
- Macgregor, H.C., and T.M. Uzzell, Jr. 1964. Gynogenesis in salamanders related to *Ambystoma jeffersonianum*. Science 143: 1043–1045.
- Morris, M.A. 1985. A hybrid *Ambystoma platineum* × *A. tigrinum* from Indiana. Herpetologica 41: 267–271.

- Niedzwiecki, J. 2005. Evolutionary history and hybridization of two mole salamander sister species from different habitats. Ph.D. thesis, University of Kentucky, Lexington.
- Petranka, J.W. 1998. Salamanders of the United States and Canada. Washington, DC: Smithsonian Institution Press.
- Ramsden, C., K. Bériault, and J.P. Bogart. 2006. A nonlethal method of identification of *Ambystoma laterale*, *A. jeffersonianum*, and sympatric unisexuals. *Molecular Ecology Notes* 6: 261–264.
- Schultz, R.J. 1969. Hybridization, unisexuality, and polyploidy in the teleost *Poeciliopsis* (Poeciliidae) and other vertebrates. *American Naturalist* 103: 605–619.
- Selander, R.K., M.H. Smith, S.Y. Yang, W.E. Johnson, and J.B. Gentry. 1971. Biochemical polymorphism and systematics in the genus *Peromyscus*. I. Variation in the Oldfield mouse (*Peromyscus polionotus*): studies in genetics IV. University of Texas Publication 7103: 49–90.
- Selander, T.C. 1994. Reproduction of unisexual hybrid *Ambystoma* (Urodela: Ambystomatidae) in the absence of parental species. M.Sc. thesis, University of Dayton, Ohio.
- Sessions, S.K. 1982. Cytogenetics of diploid and triploid salamanders of the *Ambystoma jeffersonianum* complex. *Chromosoma* 84: 599–621.
- Shaffer, H.B., and M.L. McKnight. 1996. The polytypic species revisited: genetic differentiation and molecular phylogenetics of the tiger salamander *Ambystoma tigrinum* (Amphibia: Caudata) complex. *Evolution* 50: 417–433.
- Taylor, A.S., and J.P. Bogart. 1990. Karyotypic analyses of four species of *Ambystoma* (Amphibia, Caudata) which have been implicated in the production of all-female hybrids. *Genome* 33: 837–844.
- Uzzell, T.M. 1964. Relations of the diploid and triploid species of the diploid and triploid species of the *Ambystoma jeffersonianum* complex (Amphibia, Caudata). *Copeia* 1964: 257–300.
- Uzzell, T.M., and S.M. Goldblatt. 1967. Serum proteins of salamanders of the *Ambystoma jeffersonianum* complex. *Copeia* 1967: 602–612.

APPENDIX 1

Additional Individuals Collected from Previous Collection Sites Examined by Bogart and Klemens (1997)

Localities listed by site numbers where *Ambystoma laterale* (LL), *A. jeffersonianum* (JJ), and their diploid (LJ), triploid (LLJ or LJJ), and tetraploid (LLLJ, LLJJ, LJJJ) unisexual kleptogens were identified. New sites examined (sites 107–216) continue from the 106 sites examined by Bogart and Klemens (1997). Catalogue numbers refer to specimens that are deposited in the American Museum of Natural History (AMNH) and catalogue numbers of J. P. Bogart (JPB).

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- 2) **New York:** Tompkins County: Dryden. (n = 35). JJ (AMNH 160166–200).
- 7) **New York:** Orange County: Chester, Goose Pond Mountain, Seeley Brook. (n = 4). LLJ (AMNH 164593). LLLJ (AMNH 158825, AMNH 160339–40).
- 18) **New York:** Dutchess County: Pawling, Great Swamp, South of Rte. 55 overpass. (n = 5). LL (AMNH 159999–160000). LJ (AMNH 165937). LLJ (AMNH 165994). LLJJ (AMNH 153121).
- 20) **New York:** Putnam County: Putnam Valley (n = 2). LJJ (AMNH 169808–09).
- 28) **Connecticut:** Fairfield County: Redding. (n = 1). LJJ (AMNH 158787).
- 42) **Connecticut:** Litchfield County: Bantam Lake. (n = 6). LL (AMNH 160056). LLJ (AMNH 166983–87).
- 60) **Connecticut:** Windham County: Plainfield. (n = 2). LL (AMNH 169855–56).
- 66) **Massachusetts:** Bristol and Plymouth counties: Hockomock Swamp. (n = 2). LL (AMNH 169355–56).

NEW COLLECTION SITES WEST OF THE HUDSON RIVER

New York

- 107) Orange County: Blooming Grove, Youngs Brook. (n = 25). LL (AMNH 169885–93). LJ (AMNH 169913–16). LLJ (AMNH 169900–06). LLLJ (AMNH 169899, AMNH 169909–12).
- 108) Orange County: Amity. (n = 43). LL (AMNH 160041–54). LJ (AMNH 160248–54), LLJ (AMNH 160055, AMNH 160269–84). LJJ (AMNH 153118, AMNH 160330). LLLJ (AMNH 160341–43).
- 109) Orange County: Amity Uplands. (n = 9). LJ (AMNH 169917–18). LJJ (AMNH 169849, AMNH 169918–21). LJJJ (AMNH 169922–24).
- 110) Orange County: Warwick, Wawayanda Creek – Bellvale Valley (n = 31). LL (AMNH 169369–84, AMNH 169882–84). LJ (AMNH 169395). LLJ (AMNH 165995, AMNH 169396–98, AMNH 169894–98). LLLJ (AMNH 169907–08).

APPENDIX 1
(Continued)

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- 111) Orange County: Goshen, Otter Kill floodplain swamp. (n = 26). LL (AMNH 169386–94). LLJ (AMNH 169385, AMNH 169399–410, AMNH 169806). LLLJ (AMNH 169411–13).
- 112) Orange County: Wawayanda, vicinity of Echo Lake Rd. (n = 5). LJ (AMNH 169802). LLJ (AMNH 169803–05, AMNH 169807).
- 113) Orange County: Stewart State Forest (n = 43). LL (AMNH 162962, AMNH 164587–88, AMNH 162963–65). LJ (AMNH 162819–23, AMNH 162825–30, AMNH 162946, AMNH 164590–92, AMNH 164636–38). LLJ (AMNH 162942–45, AMNH 162947–49, AMNH 162951–59). LJJ (AMNH 162939). LLLJ (AMNH 162950).
- 114) Orange County 2 mi. ESE Mount Hope. (n = 46). JJ (AMNH 153123–59, AMNH 160239–45). LJ (AMNH 153110). LJJ (AMNH 153112).
- 115) Ulster County: between Buttersville and Bonticou Crag. (n = 3). JJ (AMNH 153122). LJJ (AMNH 153116, AMNH 160337).
- 116) Ulster County: Lloyd, Black Creek floodplain. (n = 1). JJ (AMNH 169460).
- 117) Ulster County: Esopus, Black Creek Preserve. (n = 9). LJJ (AMNH 169451–55). LJJJ (AMNH 169456–59).
- 118) Otsego County: Oaks Creek drainage. (n = 16). JJ (AMNH 160223–38).
- 119) Cortland County: Scott Township. (n = 7). JJ (AMNH 160222). LJJ (AMNH 160331–36).
- 120) Tompkins County: Ithaca, Bull Pasture Pond. (n = 14). JJ (AMNH 163376–89).
- 121) Sullivan County: Tusten. (n = 20). JJ (AMNH 160202–21).
- 122) Sullivan County: Bashakill State Wildlife Management Area. (n = 7). LJ (AMNH 153107–09, AMNH 153119). LJJ (AMNH 153113–15).
- 123) Schoharie County: near South Gilboa Station along Rte. 23. (n = 14) JJ (AMNH 160201). LJ (AMNH 153111, AMNH 162832–34, AMNH 165938–44). LLJ (AMNH 165996). LJJ (AMNH 162835).
- 124) Schoharie County: Wright site. (n = 11). JJ (AMNH 169823). LJ (AMNH 169810–12). LJJ (AMNH 169816–22).
- 125) Schoharie County: Seward. (n = 5). JJ (AMNH 169824–25). LJ (AMNH 169813–14). LLJ (AMNH 169815).
- 126) Albany County: Knox. (n = 30). JJ (AMNH 169797–801). LJ (AMNH 169772–76). LJJ (AMNH 169777–94). LJJJ (AMNH 169795–96).
- 127) Seneca/Wayne Counties: Montezuma Marsh. (n = 51). LL (AMNH 160001–40, AMNH 162968). LLJ (AMNH 160257–66).
- 128) Cayuga County Mentz. (n = 1). LLJ (AMNH 160268).
- 129) Genesee County: Alabama, Orchard Swamp. (n = 12). LL (AMNH 162969–70, AMNH 165869–74). LLJ (AMNH 162961, AMNH 165997–99).
- 130) Wayne County Huron, Lummisville Road. (n = 13). JJ (AMNH 163372, AMNH 165826–30). LJ (AMNH 165945). LJJ (AMNH 162938, AMNH 165953–57).
- 131) Wayne County: Huron, Lake Ontario wetland. (n = 6). JJ (AMNH 165831). LJJ (AMNH 165958–62).
- 132) Monroe County: Wheatland, Blue Pond. (n = 2). LL (AMNH 162966). LLJ (AMNH 162960).
- 133) Orleans County: Barre, Culver Road. (n = 7). LL (AMNH 165875–77). LLJ (AMNH 166000–03).
- 134) Livingston County: Caledonia, Cement Plant Pond. (n = 2). LL (AMNH 162967). LJ (AMNH 162831).
- 135) Cattaraugus County: Mansfield. (n = 1). JJ (AMNH 163467).
- 136) Cattaraugus County: Conewango Creek. (n = 18). LL (AMNH 165878–84). LLJ (AMNH 166004–14).
- 137) Wyoming County: Arcade. (n = 12). JJ (JPB 31109, AMNH 163462–66, AMNH 165832–33, AMNH 167064). LJ (AMNH 167065–66). LJJ (AMNH 167067).
- 138) Wyoming County: Eagle, West Hill Road. (n = 4). JJ (AMNH 167068–69). LJ (AMNH 167070). LJJ (AMNH 167071).
- 139) Wyoming County: Pike, Safford Road. (n = 1). LJJ (AMNH 167072).
- 140) Wyoming County: Wethersfield. (n = 4). JJ (AMNH 169999). LJJ (AMNH 169998, AMNH 170000–01).
- 141) Wyoming County: Orangeville. (n = 3). LJJ (AMNH 170002–04).
- 142) Wyoming County: Sheldon. (n = 1). LJJ (AMNH 170005).
- 143) Erie County: Buffalo, RR yards between Tift Street and Buffalo River. (n=17). LL (AMNH 162971, AMNH 167021–32). LLJ (AMNH 167033–36).
- 144) Erie County: Grand Island. (n = 10). LL (AMNH 165885–91). LLJ (AMNH 166015–17).
- 145) Erie County: Lackawanna, South branch of Smoke Creek. (n = 19). LL (AMNH 167037–40). LLJ (AMNH 167041–55).
- 146) Erie County: Tonawanda, Kenmore. (n = 1). LL (AMNH 167056).
- 147) Niagara County: Royalton. (n = 7). LL (AMNH 167057–63).
- 148) Chautauqua County: Poland, Conewango Creek. (n = 9). LLJ (AMNH 167012–19). LLLJ (AMNH 167020).
- 149) St. Lawrence County: Lawrence. (n = 1). LLJ (AMNH 169446).

APPENDIX 1

(Continued)

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- 150) St. Lawrence County: Brasher, Bush Rd. (n = 17). LL (AMNH 169414–30).
 151) St. Lawrence County: Waddington-Louisville town line, Town Line Rd. (n = 9). LL (AMNH 169431–37). LLJ (AMNH 169447–48).
 152) St. Lawrence County: Lisbon, Swamp Rd. (n = 10). LL (AMNH 169438–45). LLJ (AMNH 169449–50).
 153) Essex County: St. Armand, Adirondack Park. (n = 1). LL (AMNH 169368).

New Jersey

- 154) Morris and Somerset Counties: Great Swamp. (n = 13). LL (AMNH 160057–68, AMNH 160071).
 155) Morris County: Troy Meadows. (n = 10). LL (AMNH 160069–70). LLJ (AMNH 160285–91). LLLJ (AMNH 160338).
 156) Morris County: Budd Lake. (n = 13). JJ (AMNH 160162–64, AMNH 163366–67, AMNH 163369). LJ (AMNH 162816–18). LJJ (AMNH 162925, AMNH 162933–35).
 157) Morris County: Stephens State Park. (n = 8). JJ (AMNH 163368). LJJ (AMNH 162926–32).
 158) Morris County: Berkshire Valley Wildlife Management Area. (n = 13). JJ (AMNH 165834–40). LJ (AMNH 165946). LJJ (AMNH 165963–67).
 159) Warren County: Hardwick Twp. (n = 9). JJ (AMNH 163370–71). LJ (AMNH 160225). LJJ (AMNH 160322–24, AMNH 162922–24).
 160) Warren County: Jenny Jump Sink. (n = 4). LJJ (AMNH 165968–71).
 161) Warren County: Frelinghuysen Twp., Glovers Pond. (n = 11). LLJ (AMNH 166999–01). LJJ (AMNH 166991–97). LJJJ (AMNH 166998).
 162) Warren County: Frelinghuysen Twp., Bear Creek. (n = 16). LL (AMNH 166988–90, AMNH 169357–58). LLJ (AMNH 167002–03, AMNH 169359–66). LLLJ (AMNH 167004).
 163) Sussex County: Swartswood. (n = 12). JJ (AMNH 163361–63). LJJ (AMNH 160329, AMNH 162890–96, AMNH 162902).
 164) Sussex County: Kittatinny Valley State Park. (n = 12). JJ (AMNH 160165, AMNH 163354–57, AMNH 162900). LJJ (AMNH 162886, AMNH 162897–99, AMNH 162901, AMNH 163554).
 165) Sussex County: Fredon Twp., Whittingham Wildlife Management Area. (n = 17). LJJ (AMNH 160321, AMNH 160325–28, AMNH 162910–21).
 166) Sussex County: Andover, north end of Stickle Pond. (n = 15). JJ (AMNH 163352–53, AMNH 163365). LJJ (AMNH 162881–85, AMNH 162903–09).
 167) Sussex County: Byram Twp., Allamuchy State Park. (n = 6). JJ (AMNH 163358–60). LJJ (AMNH 162887–89).
 168) Sussex County: Wallkill River Drainage (n = 38). LL (AMNH 160072–79, AMNH 169925). LLJ (AMNH 160292–309, AMNH 160311–17, AMNH 160320). LLLJ (AMNH 160310, AMNH 160318–19).
 169) Sussex County: Sparta Twp., Sussex County Votek Pond. (n = 7). JJ (AMNH 167005). LJ (AMNH 167006). LJJ (AMNH 167007–11).

Pennsylvania

- 170) Lehigh County: Emmaus. (n = 33). JJ (AMNH 160080–112).
 171) Lehigh County: Center Valley. (n = 10). JJ (AMNH 165841–50).
 172) Lehigh County: Salisbury Twp. (n = 9). JJ (AMNH 169483–91).
 173) Lehigh County: Upper Saucon Twp. (n = 17). JJ (AMNH 169492–506, AMNH 169929–30).
 174) Northampton County: Upper Mount Bethel Twp. (n = 3). LJ (AMNH 169928). LLJ (AMNH 169926–27).
 175) Chester County: Warwick Twp. (n = 21). JJ (AMNH 169462–82).
 176) Wyoming County: Mehoopany Twp. (n = 1). JJ (AMNH 163433).
 177) Wyoming County: Lemon Twp. (n = 5). JJ (AMNH 165851–55).
 178) Luzerne County: Lake Twp. (n = 1). JJ (AMNH 163434).
 179) Monroe County: Middle Smithfield Twp. (n = 17). JJ (AMNH 169507–10, AMNH 169834–38). LJJ (AMNH 169826–32). LJJJ (AMNH 169833).
 180) York County: near Dillsburg. (n = 25). JJ (AMNH 160113–37).
 181) Centre County: The Barrens, near Scotia. (n = 16). JJ (AMNH 160138, AMNH 163390–404).
 182) Perry County: Tuscarora State Forest. (n = 26). JJ (AMNH 163405–30).
 183) Franklin County: South Mountain. (n = 16). JJ (AMNH 163436–51).
 184) Cumberland County: Cooke Twp. (n = 2). JJ (AMNH 163431–32).
 185) Erie County: Union Twp., South Branch French Creek (n = 18). JJ (AMNH 160139–43, AMNH 167080–92).
 186) Erie County: Amity Twp., Titus Bog. (n = 18). JJ (AMNH 160144–57, AMNH 167093–96).

APPENDIX 1

(Continued)

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- 187) Erie County: Millcreek Twp. (n = 12). JJ (AMNH 163452–56, AMNH 167073–79).
 188) McKean County: Bradford, Glendorn. (n = 10). JJ (AMNH 165856–65).
 189) McKean County: Eldred. (n = 65). LL (AMNH 165892–936). LLJ (AMNH 166018–37).

Virginia

- 190) Allegheny County: 11 km. SW Covington. (n = 4). JJ (JPB 25772–75).

EAST OF THE HUDSON RIVER**Connecticut**

- 191) Fairfield County: Danbury-Ridgefield, Wooster Mountain and Mountain Pond-Eureka Lake system (n = 9). JJ (AMNH 163457, AMNH 163459–60). LJJ (AMNH 162936–37, AMNH 162940–41, AMNH 163458, AMNH 169839).
 192) Litchfield County: Washington, 0.6 mi. W Carmel Hill (n = 4). JJ (AMNH 160158). LJ (AMNH 169767). LJJ (AMNH 169768). LJJJ (AMNH 160377).
 193) Litchfield County: Norfolk, west and north slopes of Bald Mountain, 1100–1450 feet. (n = 3). LJJ (AMNH 169769–71).
 194) Litchfield County: Woodbury, Rag Land. (n = 9). JJ (AMNH 165866). LJJ (AMNH 165972–78). LJJJ (AMNH 166038).
 195) Hartford County: Granby, 0.6 mi. NW Barndoor Hills. (n = 15). JJ (AMNH 158779–84). LJJ (AMNH 158788–96).
 196) Hartford County: Simsbury, 0.2 mi. N King Phillip Mountain. (n = 3). JJ (AMNH 160159–61).
 197) Hartford County: Simsbury, Talcott Mountain. (n = 1). LLJ (AMNH 169343).
 198) Hartford County: East Granby, Marsh Pond and vicinity. (n = 13). LL (AMNH 158771, AMNH 169340–42). LLJ (AMNH 169344–45). LJJ (AMNH 158823, AMNH 169349–51). LLLJ (AMNH 169346–48).
 199) Hartford County: Farmington, Shade Swamp near Rattlesnake Mountain. (n = 1). LL (AMNH 169339).
 200) Hartford County: Farmington, 0.4 mi W Burnt Hill (n = 9). LJJ (AMNH 169840–48).
 201) Hartford County: Wethersfield, Folly Brook. (n = 7). LJ (AMNH 165947–52, AMNH 169766).
 202) Middlesex County: Durham, Pistapaug Mtn. (n = 3). LJJ (AMNH 165979–81).
 203) New Haven County: Meriden, Cathole Mtn. (n = 17). JJ (AMNH 165867). LJJ (AMNH 165982–86, AMNH 165988–93). LJJJ (AMNH 165987, AMNH 166039–42).
 204) Windham County: Plainfield and Canterbury, Quinebaug Terraces. (n = 20). LL (AMNH 169352–54, AMNH 169850–54, AMNH 169857–68).

New York

- 205) Westchester County: Lewisboro. (n = 1). JJ (AMNH 163373).
 206) Dutchess County: East Fishkill. (n = 2). LL (AMNH 169367, AMNH 169869).
 207) Dutchess County: Wingdale. (n = 12). LJJ (AMNH 169870–79). LJJJ (AMNH 169880–81).
 208) Dutchess County: Dover Furnace, Camp Sharparoon. (n = 14). LJJ (AMNH 158797–810).
 209) Dutchess County: Pawling, Swamp River below Corbin Hill. (n = 1). LLJ (AMNH 160267).
 210) Putnam County: Patterson, Muddy Brook at Cornwall Hill Road (Putnam County 64) crossing. (n = 5). LL (AMNH 158772–76).
 211) Putnam County: Great Swamp, Patterson Environmental Park. (n = 14). LL (AMNH 158777–78, AMNH 159991–98). LJ (AMNH 160246–47). LLJ (AMNH 158824, AMNH 160256).
 212) Columbia County: Ancram. (n = 2). JJ (AMNH 163374–75).
 213) Washington County: alongside Rte. 22 N of Salem, Beaver Brook drainage. (n = 1). LJJ (AMNH 153117).

Massachusetts

- 214) Middlesex County: Groton, wetlands along abandoned RR grade, S of Peabody Street, E of Groton School. (n = 4). LL (AMNH 158769). LJ (AMNH 158785–86). LLJ (AMNH 158811).
 215) Middlesex County: Groton, Groton Commons. (n = 12). LL (AMNH 158770). LJ (AMNH 158812, AMNH 158816). LLJ (AMNH 158813–15, AMNH 158817–22).
 216) Berkshire County: Sheffield, Schenob Brook drainage. (n = 1). LLJ (AMNH 153120).
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APPENDIX 2

Genotypes of *Ambystoma laterale*, *A. jeffersonianum*, and unisexual specimens ordered by site number

Genotypes of the specimens are ordered by site number (see appendix 1). Specimen numbers are American Museum of Natural History (AMNH) or catalogue numbers of J. P. Bogart (JPB). The sex of the specimen is female (F) or male (M). ?, M? or F? are juvenile individuals where the sex was difficult to determine, and L is a larval sample. The letters for the electrophoretic alleles, or allozymes, for each locus refer to relative mobilities of the stained enzyme on the gel (see table 4). A single letter is given for a homozygous condition so, for example, a B would be BB in a diploid, BBB in a triploid and BBBB in a tetraploid. Data that were not obtained for an individual at an isozyme locus is signified by —. Data for “blood” are mean erythrocyte area determinations or flow cytometric (FMC) determinations for ploidy based on comparative fluorescence of blood cells from individuals against a diploid standard fluorescence (see Ramsden et al. 2006). Blood data that were not obtained for an individual is signified by —. Tables in this appendix compare individuals that have the same genotypes:

- 2-1: *Ambystoma laterale* (LL) diploids from all sites.
- 2-2: *Ambystoma jeffersonianum* (JJ) diploids from all sites.
- 2-3: *Ambystoma laterale* – *jeffersonianum* (LJ) diploids from all sites.
- 2-4: *Ambystoma* (2) *laterale* – *jeffersonianum* (LLJ) triploids from all sites.
- 2-5: *Ambystoma laterale* – (2) *jeffersonianum* (LJJ) triploids from all sites.
- 2-6: *Ambystoma* (3) *laterale* – *jeffersonianum* (LLLJ) tetraploids from all sites.
- 2-7: *Ambystoma laterale* – (3) *jeffersonianum* (LJJJ) tetraploids from all sites.
- 2-8: *Ambystoma* (2) *laterale* – (2) *jeffersonianum* (LLJJ) tetraploid from site 18.

Appendix 2-1: Genotypes of diploid (2n) *Ambystoma laterale* specimens ordered by site number.

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
1	18	F	159999	B	B	B	B	B	D	C	—	B	B	B	704
2	18	M	160000	B	B	B	B	B	D	C	—	B	B	B	651
3	42	F	160056	B	B	B	B	B	D	—	C	BC	B	B	515
4	60	M	169855	B	B	B	B	B	D	C	C	B	B	B	FCM
5	60	F	169856	B	B	B	B	B	D	C	C	B	B	B	FCM
6	66	M	169355	B	B	B	B	A	D	C	C	B	B	B	FCM
7	66	F	169356	B	B	B	B	A	D	C	C	B	B	B	FCM
8	107	F	169885	B	B	B	B	B	D	—	C	B	B	B	FCM
9	107	F	169886	B	B	B	B	A	D	—	C	B	B	B	FCM
10)	107	M	169887	B	B	B	B	B	D	C	BC	B	B	B	FCM
11	107	F	169888	B	B	B	B	B	D	—	C	B	B	B	FCM
12	107	F	169889	B	B	B	B	A	D	—	C	B	B	B	FCM
13	107	M	169890	B	B	B	B	B	D	—	C	B	B	B	FCM
14	107	M?	169891	B	B	B	B	A	D	—	C	B	B	B	FCM
15	107	M?	169892	B	B	B	B	A	D	—	C	B	B	B	FCM
16	107	F	169893	B	B	B	B	A	D	—	C	B	B	B	FCM
17	108	M	160041	B	B	B	B	B	D	CD	—	BC	B	B	757
18	108	F	160042	B	B	B	B	B	D	C	C	B	B	B	801
19	108	F	160043	B	B	B	B	B	D	C	—	B	B	B	734
20)	108	M	160044	B	B	B	B	B	D	CD	—	B	B	B	807
21	108	M	160045	B	B	B	B	B	D	C	—	B	B	B	627
22	108	M	160046	B	B	B	B	B	D	C	—	B	B	B	770
23	108	M	160047	B	B	B	B	B	D	CD	—	B	B	B	814
24	108	M	160048	B	B	B	B	B	D	C	—	B	B	B	772
25	108	F	160049	B	B	B	B	B	D	C	—	B	B	B	772
26	108	F	160050	B	B	B	B	B	D	—	—	B	B	B	732
27	108	M	160051	B	B	B	B	A	D	CD	C	B	B	B	788
28	108	M	160052	B	B	B	B	B	D	—	C	B	B	B	709

APPENDIX 2-1
(Continued)

Locus															
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
29	108	M	160053	B	B	B	B	D	—	C	B	B	B	523	
30)	108	M	160054	B	B	B	B	A	D	—	B	B	B	800	
31	110	M	169369	B	B	B	B	D	C	—	B	AB	B	FCM	
32	110	M	169370	B	B	B	B	D	C	—	B	B	B	FCM	
33	110	F	169371	B	B	B	B	D	C	C	B	B	B	FCM	
34	110	M	169372	B	B	B	B	D	C	C	B	AB	B	FCM	
35	110	F	169373	B	B	B	B	D	C	C	B	AB	B	FCM	
36	110	M	169374	B	B	B	B	D	C	C	B	AB	B	FCM	
37	110	F	169375	B	B	B	B	D	C	C	B	AB	B	FCM	
38	110	F	169376	B	B	B	B	D	C	C	B	B	B	FCM	
39	110	F	169377	B	B	B	B	D	C	C	B	B	B	FCM	
40)	110	F	169378	B	B	B	B	D	C	C	B	AB	B	FCM	
41	110	F	169379	B	B	B	B	D	C	C	B	AB	B	FCM	
42	110	M	169380	B	B	B	B	D	C	C	B	B	B	FCM	
43	110	F	169381	B	B	B	B	D	C	C	B	B	B	FCM	
44	110	F	169382	B	B	B	B	D	C	C	B	B	B	FCM	
45	110	F	169383	B	B	B	B	D	C	C	B	AB	B	FCM	
46	110	M	169384	B	B	B	B	D	C	C	B	B	B	FCM	
47	110	F	169882	B	B	B	B	D	C	C	B	B	B	FCM	
48	110	F	169883	B	B	B	B	D	C	C	B	AB	B	FCM	
49	110	F	169884	B	B	B	B	D	C	C	B	AB	B	FCM	
50)	111	F	169386	B	B	B	B	A	D	C	—	B	B	FCM	
51	111	F	169387	B	B	B	B	A	D	C	C	B	B	FCM	
52	111	F	169388	B	B	B	B	A	D	C	—	B	B	FCM	
53	111	M	169389	B	B	B	B	A	D	—	C	B	B	FCM	
54	111	F	169390	B	B	B	B	A	D	C	—	B	B	FCM	
55	111	F	169391	B	B	B	B	A	D	C	—	B	B	FCM	
56	111	M	169392	B	B	B	B	B	D	C	C	B	B	FCM	
57	111	M	169393	B	B	B	B	A	D	C	C	B	B	FCM	
58	111	F	169394	B	B	B	B	B	D	C	C	B	B	FCM	
59	113	M	162962	B	B	B	B	B	D	C	—	B	B	691	
60)	113	F	162963	B	B	B	B	B	D	C	C	B	BC	B	
61	113	M	162964	B	B	B	B	B	D	C	C	B	B	757	
62	113	M	162965	B	B	QC	A	B	D	C	—	B	B	913	
63	113	M	164587	B	B	B	B	B	D	C	C	B	B	723	
64	113	F	164588	B	B	B	B	B	D	C	C	B	B	734	
65	127	M	160001	B	B	—	—	—	C	—	B	B	B	—	
66	127	F	160002	B	B	B	B	B	D	C	C	B	B	794	
67	127	F	160003	B	B	B	B	AB	D	C	C	B	B	700	
68	127	M	160004	B	B	B	B	A	D	C	C	B	B	705	
69	127	M	160005	B	B	B	B	A	D	C	C	B	B	671	
70)	127	M	160006	B	B	B	B	B	D	C	—	B	B	591	
71	127	F	160007	B	B	B	B	A	D	C	C	B	B	690	
72	127	M	160008	B	B	B	B	B	D	—	—	B	B	787	
73	127	M	160009	B	B	B	B	B	D	—	—	B	B	739	
74	127	F	160010	B	B	B	B	AB	D	C	C	B	B	774	
75	127	M	160011	B	B	B	B	B	D	C	—	B	B	620	
76	127	F	160012	B	B	B	B	B	D	C	—	B	B	646	
77	127	F	160013	B	B	B	B	AB	D	C	C	B	B	725	
78	127	M	160014	B	B	B	B	A	D	C	C	B	B	682	
79	127	F	160015	B	B	B	BD	B	D	C	—	B	B	714	
80)	127	M	160016	B	B	B	B	A	D	C	C	B	B	740	

APPENDIX 2-1
(Continued)

Locus														
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
81	127	M	160017	B	B	B	B	A	D	C	C	B	B	B 785
82	127	M	160018	B	B	B	B	B	D	C	C	B	B	B 692
83	127	F	160019	B	B	B	B	B	D	C	—	B	B	B 828
84	127	F	160020	B	B	B	B	AB	D	C	C	B	B	B 740
85	127	M	160021	B	B	B	B	A	D	C	C	B	B	B 778
86	127	M	160022	B	B	B	D	B	D	—	—	B	B	B 608
87	127	M	160023	B	B	B	B	B	D	C	—	B	B	B 715
88	127	F	160024	B	B	B	B	B	D	C	C	B	B	B 695
89	127	M	160025	B	B	B	B	AB	D	C	C	B	B	B 762
90)	127	M	160026	B	B	B	B	AB	D	C	C	B	B	B 661
91	127	F	160027	B	B	B	B	AB	D	C	C	B	B	B 657
92	127	F	160028	B	B	B	BD	B	D	—	—	B	B	B 756
93	127	M	160029	B	B	B	—	B	D	—	—	B	B	B 746
94	127	M	160030	B	B	B	B	B	D	C	—	B	B	B 726
95	127	F	160031	B	B	B	B	B	D	C	C	B	B	B 800
96	127	M	160032	B	B	B	B	A	D	—	—	B	B	B 687
97	127	F	160033	B	B	B	—	B	D	—	—	A	B	B 669
98	127	F	160034	B	B	B	B	B	D	—	—	A	B	B 735
99	127	F	160035	B	B	B	B	B	D	—	—	B	B	B 710
100)	127	M	160036	B	B	B	BD	B	D	—	—	B	B	B 669
101	127	M	160037	B	B	B	BD	B	D	C	—	B	B	B 640
102	127	F	160038	B	B	B	BD	BC	D	—	—	B	B	B 789
103	127	F	160039	B	B	B	B	AB	D	—	—	A	B	B 679
104	127	M	160040	B	B	B	B	B	D	—	—	B	B	B 695
105	127	M	162968	B	B	B	D	AC	D	—	B	B	B	— 949
106	129	F	162969	B	B	B	B	B	D	C	—	B	B	B 787
107	129	M	162970	B	B	B	B	B	D	C	—	B	B	B 750
108	129	F	165869	B	B	B	B	B	D	C	—	B	B	B 704
109	129	F	165870	B	B	B	B	B	D	C	—	B	B	B 708
110)	129	F	165871	B	B	B	BD	B	D	—	—	B	AB	B 688
111	129	F	165872	B	B	B	BD	B	D	C	—	B	B	B 695
112	129	F	165873	B	B	B	B	B	D	C	—	B	B	B 697
113	129	M	165874	B	B	B	B	B	D	C	—	B	B	B 744
114	132	F	162966	B	B	B	B	B	D	—	—	B	B	B 834
115	133	F	165875	B	B	B	B	B	D	C	—	B	B	B 598
116	133	F	165876	B	B	B	D	B	D	C	—	B	B	B 628
117	133	M	165877	B	B	B	B	B	D	C	—	B	B	B 717
118	134	F	162967	B	B	B	BD	B	D	—	—	B	B	B 876
119	136	F	165878	B	B	B	B	A	D	C	—	B	B	B 643
120)	136	M	165879	B	B	B	BD	B	D	C	—	B	B	B 558
121	136	M	165880	B	B	B	B	B	D	C	C	B	B	B 580
122	136	F	165881	B	B	B	B	B	D	C	—	B	B	B 593
123	136	M	165882	B	B	B	BD	B	D	C	C	B	B	B 653
124	136	M	165883	B	B	B	B	B	D	—	—	B	B	B 779
125	136	F	165884	B	B	B	B	B	D	C	C	B	B	B 648
126	143	M	162971	B	B	B	B	B	D	C	C	B	B	B 874
127	143	F	167021	B	B	B	B	A	D	C	C	B	B	B 781
128	143	F	167022	B	B	B	B	B	D	C	C	B	B	B 770
129	143	F	167023	B	B	B	B	B	D	C	C	B	B	B 772
130)	143	M	167024	B	B	B	B	B	D	C	C	B	B	B 814
131	143	M	167025	B	B	B	B	A	D	C	C	B	B	B 828
132	143	M	167026	B	B	B	BD	B	D	C	C	B	B	B 718

APPENDIX 2-1
(Continued)

Locus															
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
133	143	M	167027	B	B	B	B	D	C	C	B	B	B	764	
134	143	M	167028	B	B	B	B	D	C	C	B	B	B	773	
135	143	M	167029	B	B	B	B	D	C	C	B	B	B	793	
136	143	M	167030	B	B	B	B	D	C	C	B	B	B	811	
137	143	F	167031	B	B	B	BD	B	D	C	C	B	B	782	
138	143	F	167032	B	B	B	BD	B	D	C	C	B	B	740	
139	144	M	165885	B	B	B	B	D	—	C	B	B	B	653	
140)	144	F	165886	B	B	B	B	D	C	C	BC	B	B	728	
141	144	M	165887	B	B	B	B	D	C	—	B	B	B	700	
142	144	F	165888	B	B	B	B	D	C	—	B	B	B	687	
143	144	M	165889	B	B	B	B	D	C	—	B	B	B	686	
144	144	F	165890	B	B	B	B	D	C	—	B	B	B	696	
145	144	M	165891	B	B	B	B	D	C	—	B	B	B	802	
146	145	M	167037	B	B	B	B	A	D	C	C	B	B	733	
147	145	F	167038	B	B	B	B	D	C	C	B	B	B	705	
148	145	F	167039	B	B	B	BD	A	D	C	C	B	B	640	
149	145	M	167040	B	B	B	B	D	C	C	B	B	B	745	
150)	146	M	167056	B	B	B	B	D	C	—	B	B	B	FCM	
151	147	M	167057	B	B	B	B	D	C	—	B	B	B	FCM	
152	147	M	167058	B	B	B	B	D	C	—	B	B	B	783	
153	147	F	167059	B	B	B	B	D	C	—	B	B	B	762	
154	147	M	167060	B	B	B	B	D	C	—	B	B	B	885	
155	147	M	167061	B	B	B	B	D	C	—	B	B	B	794	
156	147	M	167062	B	B	B	B	D	C	—	B	B	B	879	
157	147	M	167063	B	B	B	B	D	C	—	B	B	B	745	
158	150	M	169414	B	B	B	B	A	CD	C	AC	B	B	FCM	
159	150	F	169415	B	B	B	B	B	D	C	—	B	B	FCM	
160)	150	F	169416	B	B	B	B	A	D	C	—	B	B	FCM	
161	150	F	169417	B	B	B	B	B	CD	C	AC	B	B	FCM	
162	150	F	169418	B	B	B	B	A	D	C	—	B	B	FCM	
163	150	F	169419	B	B	B	B	A	D	C	—	B	B	FCM	
164	150	M	169420	B	B	B	B	A	D	C	C	B	B	FCM	
165	150	F	169421	B	B	B	B	B	D	C	C	B	B	FCM	
166	150	F	169422	B	B	B	B	B	D	CD	—	B	B	FCM	
167	150	F	169423	B	B	B	B	A	D	C	—	B	B	FCM	
168	150	F	169424	B	B	B	B	B	D	C	C	B	B	FCM	
169	150	M	169425	B	B	B	B	A	D	C	—	B	B	FCM	
170)	150	F	169426	B	B	B	B	A	D	C	—	B	B	FCM	
171	150	F	169427	B	B	B	B	B	D	C	—	B	B	FCM	
172	150	F	169428	B	B	B	B	AB	D	C	—	AB	B	FCM	
173	150	M	169429	B	B	B	B	B	D	C	C	B	B	FCM	
174	150	F	169430	B	B	B	B	B	D	C	C	AB	B	FCM	
175	151	M	169431	B	B	B	B	B	D	C	C	B	B	FCM	
176	151	F	169432	B	B	B	B	A	D	C	C	B	B	FCM	
177	151	M	169433	B	B	B	B	B	D	C	C	B	B	FCM	
178	151	M	169434	B	B	B	B	B	D	C	C	B	B	FCM	
179	151	M	169435	B	B	B	B	A	D	C	—	B	B	FCM	
180)	151	F	169436	B	B	B	B	A	D	C	C	B	B	FCM	
181	151	F	169437	B	B	B	B	A	D	C	—	AB	B	FCM	
182	152	F	169438	B	B	B	B	B	D	C	C	B	B	FCM	
183	152	F	169439	B	B	B	B	AB	D	C	C	B	B	FCM	
184	152	M	169440	B	B	B	B	B	D	C	C	B	B	FCM	

APPENDIX 2-1
(Continued)

Locus															
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
185	152	F	169441	B	B	B	B	A	D	C	C	B	B	B	FCM
186	152	M	169442	B	B	B	B	B	D	C	C	B	B	B	FCM
187	152	M	169443	B	B	B	B	A	D	—	—	—	B	B	FCM
188	152	F	169444	B	B	B	B	A	D	C	—	B	B	B	FCM
189	152	M	169445	B	B	B	B	A	D	C	—	B	B	B	FCM
190)	153	F	169368	B	B	B	B	A	—	—	—	B	B	—	—
191	154	M	160057	B	B	B	B	A	D	C	—	B	B	B	699
192	154	M	160058	B	B	B	B	B	D	C	—	B	B	B	671
193	154	M	160059	B	B	B	B	A	D	C	—	B	B	B	642
194	154	M	160060	B	B	B	B	B	D	C	—	B	B	B	656
195	154	M	160061	B	B	B	B	B	D	C	—	B	B	B	780
196	154	M	160062	B	B	B	B	B	D	C	—	B	B	B	691
197	154	F	160063	B	B	B	B	AB	D	C	C	B	B	B	621
198	154	F	160064	B	B	B	B	B	D	C	C	B	B	B	657
199	154	F	160065	B	B	B	B	B	D	C	C	B	B	B	666
200)	154	F	160066	B	B	B	B	B	D	C	C	B	B	B	664
201	154	M	160067	B	B	B	B	B	D	C	C	B	B	B	686
202	154	F	160068	B	B	B	B	B	D	C	C	B	B	B	596
203	154	M	160071	B	B	B	B	B	D	C	—	B	B	B	641
204	155	F	160069	B	B	B	B	A	D	C	—	B	B	B	618
205	155	M	160070	B	B	B	B	A	D	C	C	B	B	B	698
206	162	M	166988	B	B	B	B	B	D	C	C	BC	B	B	739
207	162	F	166989	B	B	B	B	B	D	C	C	B	B	B	802
208	162	M	166990	B	B	B	B	B	D	C	—	B	B	B	FCM
209	162	M	169357	B	B	B	B	B	D	C	C	B	B	B	FCM
210)	162	M	169358	B	B	B	B	B	D	C	C	B	B	B	FCM
211	168	F	160072	B	B	B	B	A	D	C	C	B	B	B	790
212	168	M	160073	B	B	B	B	A	D	C	—	B	B	B	653
213	168	F	160074	B	B	B	B	A	D	—	C	B	B	B	572
214	168	M	160075	B	B	B	B	A	D	—	—	B	B	B	742
215	168	F	160076	B	B	B	B	A	D	C	C	B	B	B	680
216	168	F	160077	B	B	B	B	B	D	—	C	B	B	B	698
217	168	M	160078	B	B	B	B	A	D	C	—	B	B	B	769
218	168	F	160079	B	B	B	B	A	D	C	—	B	B	B	779
219	168	M	169925	B	B	B	B	B	D	C	AC	B	B	B	FCM
220)	189	M	165892	B	B	B	BD	B	D	C	C	B	B	B	610
221	189	M	165893	B	B	B	D	B	D	C	C	B	B	B	693
222	189	M	165894	B	B	B	BD	A	D	C	C	B	B	B	690
223	189	F	165895	B	B	B	BD	B	D	C	C	B	B	B	813
224	189	F	165896	B	B	B	BD	A	D	C	C	C	B	B	752
225	189	F	165897	B	B	B	BD	B	D	C	C	BC	B	B	655
226	189	M	165898	B	B	B	D	B	D	C	C	B	B	B	810
227	189	F	165899	B	B	B	BD	B	D	C	C	B	B	B	604
228	189	F	165900	B	B	B	B	B	D	C	C	B	B	B	591
229	189	F	165901	B	B	B	BD	B	D	C	C	B	B	B	658
230)	189	M	165902	B	B	B	B	B	D	C	C	B	B	B	669
231	189	M	165903	B	B	B	BD	B	D	C	C	B	B	B	613
232	189	M	165904	B	B	B	BD	A	D	C	C	B	B	B	670
233	189	F	165905	B	B	B	D	B	D	C	C	B	B	B	665
234	189	F	165906	B	B	B	BD	B	D	C	C	B	B	B	721
235	189	M	165907	B	B	B	B	B	D	C	C	B	B	B	—
236	189	F	165908	B	B	B	B	B	D	C	C	B	B	B	891

APPENDIX 2-1
(Continued)

				Locus											
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
237	189	M	165909	B	B	B	D	B	D	C	C	B	B	B	865
238	189	F	165910	B	B	B	BD	B	D	C	—	B	B	B	721
239	189	M	165911	B	B	B	B	B	D	C	—	B	B	B	620
240)	189	M	165912	B	B	B	BD	B	D	C	—	B	B	B	694
241	189	F	165913	B	B	B	D	B	D	C	—	B	B	B	613
242	189	F	165914	AB	B	B	BD	B	D	C	—	B	B	B	700
243	189	M	165915	B	B	B	D	B	D	C	—	B	B	B	654
244	189	M	165916	B	B	B	D	B	D	C	—	B	B	B	656
245	189	M	165917	AB	BC	B	BD	B	D	C	—	B	B	B	592
246	189	F	165918	B	B	B	B	B	D	C	—	B	B	B	798
247	189	M	165919	B	C	B	B	B	D	C	—	B	B	B	651
248	189	F	165920	B	B	B	B	B	D	C	—	B	B	B	598
249	189	F	165921	B	B	B	BD	B	D	C	—	B	B	B	620
250)	189	F	165922	B	B	B	BD	B	D	C	—	B	B	B	641
251	189	M	165923	B	B	B	B	B	D	C	—	B	B	B	694
252	189	F	165924	B	B	B	BD	B	D	C	—	B	B	B	782
253	189	M	165925	B	B	B	D	B	D	C	—	B	B	B	793
254	189	M	165926	B	B	B	B	B	D	C	—	B	B	B	686
255	189	F	165927	B	B	B	D	B	D	C	—	B	B	B	724
256	189	M	165928	B	B	B	D	B	D	C	—	B	B	B	803
257	189	M	165929	B	B	B	D	B	D	C	—	B	B	B	730
258	189	M	165930	B	B	B	D	B	D	C	—	B	B	B	624
259	189	M	165931	B	B	B	B	B	D	C	—	B	B	B	657
260)	189	F	165932	B	B	B	D	B	D	C	—	B	B	B	698
261	189	M	165933	B	B	B	D	B	D	C	—	B	B	B	660
262	189	F	165934	B	B	B	BD	B	D	C	—	B	B	B	586
263	189	M	165935	B	B	QB	D	AB	D	C	—	B	B	B	790
264	189	F	165936	B	B	B	D	B	D	C	—	B	B	B	650
265	198	F	158771	B	B	B	B	B	D	C	—	B	B	B	788
266	198	M	169340	B	B	B	B	B	D	C	—	B	B	B	FCM
267	198	M	169341	B	B	B	B	B	D	C	C	B	B	B	FCM
268	198	M	169342	B	B	B	B	B	CD	C	C	B	B	B	FCM
269	199	F	169339	B	B	B	—	—	C	—	—	—	B	B	—
270)	204	M	169352	B	B	B	B	B	D	C	C	B	B	B	FCM
271	204	M	169353	B	B	B	B	B	D	C	—	B	B	B	—
272	204	F	169354	B	B	B	B	B	D	—	—	B	B	B	FCM
273	204	F	169850	B	B	B	B	B	D	C	C	B	B	B	FCM
274	204	F	169851	B	B	B	B	B	D	C	C	B	B	B	FCM
275	204	F	169852	B	B	B	B	B	D	C	C	B	B	B	FCM
276	204	F	169853	B	B	B	B	B	D	C	C	B	B	B	FCM
277	204	M	169854	B	B	B	B	B	D	C	AC	B	B	B	FCM
278	204	M	169857	B	B	B	B	B	D	C	A	B	B	B	FCM
279	204	M	169858	B	B	B	B	B	D	C	C	B	B	B	FCM
280)	204	M	169859	B	B	B	B	B	D	C	C	B	B	B	FCM
281	204	M	169860	B	B	B	B	A	D	C	C	B	B	B	FCM
282	204	M	169861	B	B	B	B	A	D	C	A	B	B	B	FCM
283	204	M	169862	B	B	B	B	A	D	C	C	B	B	B	FCM
284	204	M	169863	B	B	B	B	B	D	C	C	B	B	B	FCM
285	204	M	169864	B	B	B	B	A	D	C	C	B	B	B	FCM
286	204	M	169865	B	B	B	B	B	D	C	AC	B	B	B	FCM
287	204	M	169866	B	B	B	B	B	D	C	A	B	B	B	FCM
288	204	M	169867	B	B	B	B	B	D	C	A	B	B	B	FCM

APPENDIX 2-1
(Continued)

	Site	Sex	AMNH	Locus											
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
289	204	F	169868	B	B	B	B	A	D	C	C	B	B	B	FCM
290)	206	M	169367	B	B	B	BC	B	D	C	C	B	B	B	FCM
291	206	F	169869	B	B	B	B	B	D	C	C	B	B	B	FCM
292	210	F	158772	B	B	B	B	B	D	C	C	B	B	B	605
293	210	M	158773	B	B	B	B	B	D	C	C	B	B	B	671
294	210	M	158774	B	B	B	B	B	D	—	C	B	B	B	729
295	210	F	158775	B	B	B	—	B	D	—	—	B	B	B	728
296	210	M	158776	B	B	B	B	B	D	—	—	B	B	B	728
297	211	F	158777	B	B	B	—	B	D	—	—	B	B	B	650
298	211	M	158778	B	B	B	B	B	D	—	—	B	B	B	622
299	211	M	159991	B	B	B	B	B	D	C	—	B	B	B	683
300)	211	M	159992	B	B	B	B	B	D	C	—	B	B	B	713
301	211	M	159993	B	B	B	B	B	D	C	—	B	B	B	721
302	211	M	159994	B	B	B	B	B	D	C	—	B	B	B	661
303	211	M	159995	B	B	B	B	B	D	C	—	B	B	B	708
304	211	M	159996	B	B	B	B	B	D	C	—	B	B	B	724
305	211	M	159997	B	B	B	B	B	D	C	—	B	B	B	723
306	211	M	159998	B	B	B	B	B	D	C	—	B	B	B	782
307	214	F	158769	B	B	B	B	B	D	C	—	B	B	B	689
308	215	M	158770	B	B	B	B	B	D	C	C	B	B	B	750

Appendix 2-2: Genotypes of diploid (2n) *Ambystoma jeffersonianum* specimens ordered by site number.

	Site	Sex	AMNH	Locus											
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
1	2	M	160166	D	A	A	C	B	B	BC	AB	B	B	D	627
2	2	M	160167	D	A	A	C	B	B	B	A	C	B	D	798
3	2	M	160168	D	A	A	C	B	B	BC	—	C	B	D	776
4	2	F	160169	D	A	A	C	B	B	BC	—	BC	B	D	814
5	2	M	160170	D	A	A	C	B	BD	BC	—	B	B	D	802
6	2	M	160171	D	A	A	C	B	D	—	AB	C	B	D	628
7	2	M	160172	D	A	A	C	B	D	—	AB	BC	B	D	731
8	2	M	160173	D	A	A	C	B	B	—	A	BC	B	D	783
9	2	M	160174	D	A	A	C	B	B	BC	—	BC	B	D	719
10)	2	M	160175	D	A	A	C	B	BD	BC	—	B	B	D	693
11	2	F	160176	D	A	A	C	B	BD	—	A	BC	B	D	558
12	2	M	160177	D	A	A	C	BQ	BD	—	AB	C	B	D	747
13	2	M	160178	D	A	A	C	B	BD	—	AB	C	B	D	840
14	2	M	160179	D	A	A	C	BQ	B	—	AB	BC	B	D	790
15	2	M	160180	D	A	A	C	B	BD	—	B	BC	B	D	776
16	2	M	160181	D	A	A	C	B	BD	—	B	BC	B	D	810
17	2	M	160182	D	A	A	C	B	BD	—	AB	B	B	D	789
18	2	F	160183	D	A	A	C	B	B	—	AB	B	B	D	742
19	2	M	160184	D	A	A	C	B	B	—	A	B	B	D	717
20)	2	M	160185	D	A	A	C	B	B	—	A	BC	B	D	801
21	2	F	160186	D	A	A	C	B	BD	—	AB	BC	B	D	758
22	2	M	160187	D	A	A	C	B	D	—	AB	B	B	D	774

APPENDIX 2-2
(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
23	2	M	160188	D	A	A	C	B	BD	—	A	BC	B	D	777
24	2	F	160189	D	A	A	C	B	B	—	AB	B	B	D	759
25	2	M	160190	D	A	A	C	B	B	BC	—	BC	B	D	807
26	2	F	160191	D	A	A	C	B	B	BC	—	C	B	D	723
27	2	M	160192	D	A	A	C	B	BD	B	—	B	B	D	780
28	2	F	160193	D	A	A	C	B	B	B	—	C	B	D	772
29	2	F	160194	D	A	A	C	B	BD	B	—	C	B	D	837
30)	2	M	160195	D	A	A	C	B	BD	B	—	BC	B	D	768
31	2	F	160196	D	A	A	C	B	B	BC	—	BC	B	D	650
32	2	M	160197	D	A	A	C	B	B	BC	—	BC	B	D	744
33	2	M	160198	D	A	A	C	B	BD	B	—	BC	B	D	864
34	2	M	160199	D	A	A	C	B	BD	B	—	BC	B	D	816
35	2	F	160200	D	A	A	C	B	B	BC	—	C	B	D	—
36	114	F	153123	D	A	A	C	B	B	B	—	BC	B	D	660
37	114	F	153124	D	A	A	C	B	B	—	—	C	B	D	794
38	114	M	153125	D	A	A	C	B	B	—	—	C	B	D	602
39	114	M	153126	D	A	A	C	B	B	—	—	BC	B	D	704
40)	114	M	153127	D	A	A	C	B	B	—	—	BC	B	D	683
41	114	M	153128	D	A	A	C	B	B	B	—	B	B	D	848
42	114	F	153129	D	A	A	C	B	B	B	—	BC	B	D	805
43	114	M	153130	D	A	A	C	B	B	B	—	BC	B	D	785
44	114	M	153131	D	A	A	C	B	B	B	—	BC	B	D	701
45	114	F	153132	D	A	A	C	B	B	—	—	BC	B	D	720
46	114	F	153133	D	A	A	C	B	B	—	—	B	B	D	672
47	114	M	153134	D	A	A	C	B	B	—	—	B	B	D	656
48	114	F	153135	D	A	A	C	B	B	—	—	B	B	D	731
49	114	F	153136	D	A	A	C	B	B	B	—	C	B	D	753
50)	114	M	153137	D	A	A	C	B	B	B	—	BC	B	D	668
51	114	F	153138	D	A	A	C	B	B	B	—	C	B	D	718
52	114	M	153139	D	A	A	C	B	B	B	—	BC	B	D	785
53	114	F	153140	D	A	A	C	B	B	—	—	BC	B	D	682
54	114	F	153141	D	A	A	C	B	B	—	—	B	B	D	665
55	114	F	153142	D	A	A	C	B	B	—	—	C	B	D	680
56	114	F	153143	D	A	A	C	B	B	—	—	C	B	D	749
57	114	M	153144	D	A	A	C	B	B	—	—	C	B	D	708
58	114	F	153145	D	A	A	C	B	B	B	A	C	B	D	695
59	114	M	153146	D	A	A	C	B	B	B	—	B	B	D	724
60)	114	F	153147	D	A	A	C	B	B	—	—	B	B	D	670
61	114	F	153148	D	A	A	C	B	B	—	—	BC	B	D	654
62	114	M	153149	D	A	A	C	B	B	—	A	BC	B	D	591
63	114	M	153150	D	A	A	C	B	B	—	A	BC	B	D	716
64	114	F	153151	D	A	A	C	B	B	—	A	BC	B	D	668
65	114	M	153152	D	A	A	C	B	B	B	—	BC	B	D	788
66	114	F	153153	D	A	A	C	B	B	B	—	C	B	D	732
67	114	F	153154	D	A	A	C	B	B	—	—	BC	B	D	767
68	114	?	153155	D	A	A	C	B	B	—	—	C	B	D	669
69	114	F	153156	D	A	A	C	B	B	B	—	BC	B	D	766
70)	114	M	153157	D	A	A	C	B	B	—	—	BC	B	D	589
71	114	F	153158	D	A	A	C	B	B	—	—	C	B	D	662
72	114	M	153159	D	A	A	C	B	B	—	B	BC	B	D	641
73	114	F	160239	D	A	A	C	B	B	B	B	C	B	D	723
74	114	M	160240	D	A	A	C	B	B	B	—	C	B	D	667

APPENDIX 2-2
(Continued)

				Locus												
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
75	114	M	160241	D	A	A	C	B	B	B	—	BC	B	D	803	
76	114	M	160242	D	A	A	C	B	B	B	—	BC	B	D	765	
77	114	F	160243	D	A	A	C	B	B	B	—	C	B	D	700	
78	114	F	160244	D	A	A	C	B	B	B	—	B	B	D	717	
79	114	F	160245	D	A	A	C	B	B	B	—	BC	B	D	805	
80)	115	F	153122	D	A	A	C	B	B	—	AB	C	B	D	675	
81	116	F	169460	D	A	A	C	B	B	B	—	BC	B	D	FCM	
82	118	M	160223	D	A	A	C	B	B	—	B	C	B	D	743	
83	118	M	160224	D	A	A	C	B	BD	—	AB	C	B	D	784	
84	118	M	160225	D	A	A	C	B	BD	—	QA	C	B	D	699	
85	118	M	160226	D	A	A	C	B	B	—	B	C	B	D	740	
86	118	M	160227	D	A	QA	C	B	BD	—	AB	C	B	D	824	
87	118	F	160228	D	—	A	C	B	B	—	AB	B	B	D	652	
88	118	M	160229	D	A	A	C	B	BD	—	—	C	B	D	737	
89	118	F	160230	D	A	A	C	B	BD	—	—	C	B	D	676	
90)	118	M	160231	D	A	A	C	B	D	—	AB	C	B	D	652	
91	118	F	160232	D	A	A	C	B	BD	—	AB	C	B	D	712	
92	118	F	160233	D	A	A	C	B	BD	—	AB	C	B	D	708	
93	118	M	160234	D	A	A	C	B	D	B	AB	C	B	D	709	
94	118	M	160235	D	A	A	C	B	BD	—	AB	BC	B	D	896	
95	118	F	160236	D	A	A	C	B	D	—	AB	C	B	D	712	
96	118	M	160237	D	A	A	C	B	B	—	B	C	B	D	704	
97	118	F	160238	D	A	A	C	B	B	—	AB	C	B	D	728	
98	119	F	160222	D	A	A	C	B	BD	BC	—	C	B	D	945	
99	120	M	163376	D	A	A	C	B	BD	BC	AB	C	B	D	780	
100)	120	F	163377	D	A	A	C	B	D	B	A	C	B	D	724	
101	120	F	163378	D	A	A	C	B	D	B	B	BC	B	D	686	
102	120	M	163379	D	A	A	C	B	BD	BC	AB	BC	B	D	687	
103	120	F	163380	D	A	A	C	BQ	BD	B	AB	B	B	D	725	
104	120	F	163381	D	A	A	C	B	BD	—	—	BC	B	D	678	
105	120	F	163382	D	A	A	C	B	BD	B	B	BC	B	D	852	
106	120	F	163383	D	A	A	C	B	B	B	AB	C	B	D	798	
107	120	F	163384	D	A	A	C	B	BD	BC	—	C	B	D	699	
108	120	F	163385	D	A	A	C	B	B	B	AB	C	B	D	769	
109	120	M	163386	D	A	A	C	B	BD	B	B	B	B	D	869	
110)	120	F	163387	D	A	A	C	B	B	BC	—	C	B	D	774	
111	120	M	163388	D	A	A	C	B	B	—	A	BC	B	D	781	
112	120	M	163389	D	A	A	C	B	BD	BC	AB	C	B	D	734	
113	121	M	160202	D	A	A	C	B	B	B	AB	BC	B	D	674	
114	121	M	160203	D	A	A	C	B	B	B	AB	C	B	D	626	
115	121	M	160204	D	A	A	C	B	B	B	—	C	B	D	662	
116	121	M	160205	D	A	A	C	B	B	B	AB	BC	B	D	698	
117	121	M	160206	D	A	A	C	B	B	B	A	BC	B	D	746	
118	121	M	160207	D	A	A	C	B	B	B	AB	BC	B	D	753	
119	121	M	160208	D	A	A	C	B	B	B	AB	C	B	D	700	
120)	121	M	160209	D	A	A	C	B	B	B	A	C	B	D	554	
121	121	F	160210	D	A	A	C	B	B	—	AB	C	B	D	—	
122	121	F	160211	D	A	A	C	B	B	B	—	BC	B	D	—	
123	121	M	160212	D	A	A	C	B	B	B	—	C	B	D	—	
124	121	F	160213	D	A	A	C	B	B	B	—	AC	B	D	725	
125	121	M	160214	D	A	A	C	B	B	B	—	BC	B	D	725	
126	121	M	160215	D	A	A	C	B	B	B	—	C	B	D	853	

APPENDIX 2-2
(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
127	121	F	160216	D	A	A	C	B	B	B	—	C	B	D	617
128	121	F	160217	D	A	A	C	B	B	B	—	BC	B	D	773
129	121	M	160218	D	A	A	C	B	B	B	—	C	B	D	835
130)	121	M	160219	D	A	A	C	B	B	B	—	BC	B	D	734
131	121	F	160220	D	A	A	C	B	B	B	—	BC	B	D	726
132	121	F	160221	D	A	A	C	B	B	B	—	B	B	D	734
133	123	M	160201	D	A	A	C	B	B	—	A	C	B	D	747
134	124	F	169823	D	A	A	C	B	B	B	A	C	B	D	FCM
135	125	F	169824	D	A	A	C	B	B	B	AB	C	B	D	FCM
136	125	F	169825	D	A	A	C	B	B	B	—	C	B	D	FCM
137	126	F	169797	D	A	A	C	B	B	B	—	C	B	D	FCM
138	126	F	169798	D	A	A	C	B	B	B	A	B	B	D	FCM
139	126	M	169799	D	A	A	C	B	B	B	A	BC	B	D	FCM
140)	126	M	169800	D	A	A	C	B	B	A	—	BC	B	D	FCM
141	126	M	169801	D	A	A	C	B	B	B	A	BC	B	D	FCM
142	130	F	163372	D	A	A	C	B	B	B	AB	B	B	D	860
143	130	F	165826	D	A	A	C	B	B	B	C	C	B	D	813
144	130	F	165827	D	A	A	C	B	BD	B	—	C	B	D	853
145	130	F	165828	D	A	A	C	B	B	B	C	C	B	D	754
146	130	F	165829	D	A	A	C	B	B	B	AC	C	B	D	748
147	130	M	165830	D	A	A	C	B	B	B	AB	C	B	D	796
148	131	M	165831	D	A	A	C	B	B	B	—	C	B	D	723
149	135	M	163467	D	A	A	AC	B	B	B	AB	C	B	D	—
150)	137	?	JPB 31109	D	A	A	C	B	B	B	A	—	B	D	—
151	137	M	163462	D	A	A	C	B	B	B	A	C	B	D	742
152	137	M	163463	D	A	A	C	B	B	B	A	C	B	D	711
153	137	F	163464	D	A	A	C	B	B	B	A	C	B	D	696
154	137	M	163465	D	A	A	C	B	B	B	A	C	B	D	769
155	137	M	163466	D	A	A	C	B	B	B	A	C	B	D	822
156	137	M	165832	D	A	A	C	B	B	B	—	C	B	D	784
157	137	F	165833	D	A	A	C	B	B	B	—	C	B	D	705
158	137	M	167064	D	A	A	C	B	B	B	—	C	B	D	764
159	138	F	167068	D	A	A	C	B	B	BD	—	—	B	D	788
160)	138	M	167069	D	A	A	C	B	B	B	AB	B	B	D	783
161	140	M	169999	D	A	A	C	B	B	B	—	B	B	D	FMC
162	156	M	160162	D	A	A	C	B	B	—	—	BC	B	D	706
163	156	M	160163	D	AB	A	C	B	B	—	—	C	B	D	674
164	156	M	160164	D	A	A	C	B	B	—	—	BC	B	D	639
165	156	M	163366	D	A	A	C	B	B	B	—	C	B	D	688
166	156	M	163367	D	A	A	C	B	B	B	—	BC	B	D	798
167	156	F	163369	D	A	A	C	B	B	B	AB	B	B	D	761
168	157	M	163368	D	AC	A	C	B	B	—	AB	BC	B	D	832
169	158	M	165834	D	A	A	C	B	B	—	—	BC	B	D	790
170)	158	M	165835	D	A	A	C	B	B	B	—	C	B	D	710
171	158	M	165836	D	A	A	—	B	AB	B	—	BC	B	D	387
172	158	F	165837	D	A	A	AC	B	AB	B	—	B	B	D	476
173	158	M	165838	D	A	A	C	B	B	B	AB	B	B	D	—
174	158	M	165839	D	A	A	C	B	B	B	—	B	B	D	804
175	158	M	165840	D	A	A	C	B	B	B	—	BC	B	D	823
176	159	M	163370	D	A	A	C	B	B	B	A	BC	B	D	718
177	159	F	163371	D	A	A	C	B	B	B	A	C	B	D	766
178	163	M	163361	D	A	A	C	B	B	B	—	BC	B	D	749

APPENDIX 2-2
(Continued)

				Locus											Sod-1	blood
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2			
179	163	M	163362	D	A	A	C	B	B	B	—	C	B	D	712	
180)	163	F	163363	D	A	A	C	B	B	B	A	C	B	D	849	
181	164	M	160165	D	A	A	C	B	B	B	A	BC	B	D	785	
182	164	F	162900	D	B	A	C	B	—	—	A	—	—	D	728	
183	164	M	163354	D	A	A	C	B	B	B	AB	BC	B	D	803	
184	164	M	163355	D	A	A	C	B	B	B	—	BC	B	D	781	
185	164	M	163356	D	A	A	C	B	B	B	—	C	B	D	810	
186	164	M	163357	D	A	A	C	B	B	B	—	BC	B	D	769	
187	166	M	163352	D	A	A	C	B	B	B	A	BC	B	D	907	
188	166	M	163353	D	A	A	C	B	B	B	—	BC	B	D	830	
189	166	F	163365	D	A	A	C	B	B	—	A	C	B	D	933	
190)	167	M	163358	D	A	A	C	B	B	B	A	B	B	D	817	
191	167	F	163359	D	A	A	C	B	B	B	A	BC	B	D	919	
192	167	M	163360	D	AC	A	C	B	B	B	A	B	B	D	837	
193	169	M	167005	D	A	A	C	B	B	B	—	BC	B	D	728	
194	170	M	160080	D	A	A	C	B	B	B	—	B	B	D	710	
195	170	M	160081	D	A	A	C	B	B	B	—	B	B	D	590	
196	170	M	160082	D	A	A	C	B	B	B	—	B	B	D	721	
197	170	M	160083	D	A	A	C	B	B	B	A	C	B	D	670	
198	170	M	160084	D	A	A	C	B	B	B	A	BC	B	D	756	
199	170	M	160085	D	A	A	C	B	B	B	AB	B	B	D	683	
200)	170	F	160086	D	A	A	C	B	B	B	A	B	B	D	628	
201	170	M	160087	B	A	A	C	B	B	B	—	B	B	D	693	
202	170	M	160088	D	A	A	C	B	B	B	—	B	B	D	776	
203	170	M	160089	D	A	A	C	B	B	B	—	B	B	D	662	
204	170	M	160090	D	A	A	C	B	B	B	—	C	B	D	717	
205	170	M	160091	D	A	A	C	B	B	B	—	BC	B	D	738	
206	170	M	160092	D	A	A	C	B	B	B	—	C	B	D	681	
207	170	M	160093	D	A	A	C	B	B	B	—	B	B	D	734	
208	170	M	160094	D	A	A	C	B	B	B	—	BC	B	D	702	
209	170	M	160095	D	A	A	C	B	B	—	—	B	B	D	735	
210)	170	M	160096	D	A	A	C	B	B	—	A	BC	B	D	769	
211	170	M	160097	D	A	A	C	B	B	—	A	B	B	D	706	
212	170	M	160098	D	A	A	C	B	B	B	QA	BC	B	D	778	
213	170	M	160099	D	A	A	C	B	B	B	A	BC	B	D	663	
214	170	F	160100	D	A	A	C	B	B	B	—	BC	B	D	794	
215	170	M	160101	D	A	A	C	B	B	B	—	B	B	D	799	
216	170	F	160102	D	A	A	C	B	B	—	A	B	B	D	753	
217	170	M	160103	D	A	A	C	B	B	—	—	B	B	D	703	
218	170	F	160104	D	A	A	C	B	B	—	—	B	B	D	770	
219	170	M	160105	D	A	A	C	B	B	—	—	B	B	D	705	
220)	170	F	160106	D	A	A	C	B	B	—	A	B	B	D	692	
221	170	F	160107	D	A	A	C	B	B	—	AB	B	B	D	740	
222	170	M	160108	D	A	A	C	B	B	—	A	BC	B	D	793	
223	170	F	160109	D	A	A	C	B	B	—	A	B	B	D	649	
224	170	F	160110	D	A	A	C	B	B	—	A	BC	B	D	658	
225	170	M	160111	D	A	A	C	B	B	—	—	B	B	D	732	
226	170	M	160112	D	A	A	C	B	B	—	—	BC	B	D	722	
227	171	M	165841	D	A	A	C	B	B	B	AB	B	B	D	877	
228	171	M	165842	D	A	A	C	B	B	—	A	B	B	D	778	
229	171	F	165843	D	A	A	C	B	B	—	A	B	B	D	733	
230)	171	M	165844	D	A	A	C	B	B	—	A	B	B	D	802	

APPENDIX 2-2
(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
231	171	M	165845	D	A	A	C	B	B	—	AC	—	B	D	838
232	171	M	165846	D	A	A	C	B	B	B	A	BC	B	D	795
233	171	M	165847	D	A	A	C	B	B	—	A	B	B	D	713
234	171	F	165848	D	A	A	C	B	B	—	AB	C	B	D	699
235	171	M	165849	D	A	A	C	B	B	—	A	BC	B	D	765
236	171	M	165850	D	A	A	C	B	B	—	AC	B	B	D	788
237	172	M	169483	D	A	A	C	B	B	B	B	B	B	D	FCM
238	172	M	169484	D	A	A	C	B	B	B	A	B	B	D	FCM
239	172	M	169485	D	A	A	C	B	B	B	B	B	B	D	FCM
240)	172	M	169486	D	A	A	C	B	B	B	A	B	B	D	FCM
241	172	M	169487	D	A	A	C	B	B	B	A	B	B	D	FCM
242	172	F	169488	D	A	A	C	B	B	B	AB	B	B	D	FCM
243	172	F	169489	D	A	A	C	B	B	B	A	B	B	D	FCM
244	172	F	169490	D	A	A	C	B	B	B	AB	B	B	D	FCM
245	172	F	169491	D	A	A	C	B	B	B	A	B	B	D	FCM
246	173	M	169492	D	A	A	C	B	B	B	A	B	B	D	FCM
247	173	M	169493	D	A	A	C	B	B	B	A	B	B	D	FCM
248	173	M	169494	D	A	A	C	B	B	B	A	B	B	D	FCM
249	173	M	169495	D	A	A	C	B	B	B	A	B	B	D	FCM
250)	173	M	169496	D	A	A	C	B	B	B	A	C	B	D	FCM
251	173	M	169497	D	A	A	C	B	B	B	A	—	B	D	FCM
252	173	M	169498	D	A	A	C	B	B	B	—	B	B	D	FCM
253	173	M	169499	D	A	A	C	B	B	B	A	B	B	D	FCM
254	173	M	169500	D	A	A	C	B	B	B	A	B	B	D	FCM
255	173	M	169501	D	A	A	C	B	B	B	A	B	B	D	FCM
256	173	F	169502	D	A	A	C	B	B	B	A	B	B	D	FCM
257	173	F	169503	D	A	A	C	B	B	B	AC	C	B	D	FCM
258	173	F	169504	D	A	A	C	B	B	B	A	B	B	D	FCM
259	173	F	169505	D	A	A	C	B	B	B	A	B	B	D	FCM
260)	173	F	169506	D	A	A	C	B	B	B	A	B	B	D	FCM
261	173	F	169929	D	A	A	C	B	B	B	A	B	B	D	FCM
262	173	F	169930	D	A	A	C	B	B	B	A	B	B	D	FCM
263	175	M	169462	D	A	A	C	B	B	B	AB	BC	B	D	FCM
264	175	M	169463	D	A	A	C	B	B	B	—	BC	B	D	FCM
265	175	M	169464	D	A	A	C	B	B	B	B	BC	B	D	FCM
266	175	M	169465	D	A	A	C	B	B	B	A	BC	B	D	FCM
267	175	M	169466	D	A	A	C	B	B	B	AB	B	B	D	FCM
268	175	M	169467	D	A	A	C	B	B	B	B	B	B	D	FCM
269	175	M	169468	D	A	A	C	B	B	B	AC	B	B	D	FCM
270)	175	M	169469	D	A	A	C	B	B	B	AB	B	B	D	FCM
271	175	M	169470	D	A	A	C	B	B	B	A	C	B	D	FCM
272	175	M	169471	D	A	A	C	B	B	B	—	BC	B	D	FCM
273	175	M	169472	D	A	A	C	B	B	B	B	B	B	D	FCM
274	175	M	169473	D	A	A	C	B	B	B	AB	C	B	D	FCM
275	175	M	169474	D	A	A	C	B	B	B	B	C	B	D	FCM
276	175	M	169475	D	A	A	C	B	B	B	B	C	B	D	FCM
277	175	M	169476	D	A	A	C	B	B	B	B	B	B	D	FCM
278	175	F	169477	D	A	A	C	B	B	B	B	B	B	D	FCM
279	175	F	169478	D	A	A	C	B	B	B	B	BC	B	D	FCM
280)	175	F	169479	D	A	A	C	B	B	B	B	B	B	D	FCM
281	175	F	169480	D	A	A	C	B	B	B	B	BC	B	D	FCM
282	175	F	169481	D	A	A	C	B	B	B	AB	B	B	D	FCM

APPENDIX 2-2
(Continued)

				Locus											
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
283	175	F	169482	D	A	A	C	B	B	B	AB	B	B	D	FCM
284	176	F	163433	D	A	A	C	B	B	B	AB	B	B	D	794
285	177	M	165851	D	A	A	C	B	B	—	AB	C	B	D	725
286	177	M	165852	D	A	A	C	B	B	—	A	BC	B	D	837
287	177	M	165853	D	A	A	C	B	B	B	—	C	B	D	754
288	177	M	165854	D	A	A	C	B	B	—	—	C	B	D	761
289	177	M	165855	D	A	A	C	B	B	B	—	C	B	D	722
290)	178	F	163434	D	A	A	C	B	B	B	A	C	B	D	891
291	179	M	169507	D	A	A	C	B	B	B	AB	BC	B	D	FCM
292	179	M	169508	D	A	A	C	B	B	B	—	BC	B	D	FCM
293	179	M	169509	D	A	A	C	B	B	B	—	BC	B	D	FCM
294	179	F	169510	D	A	A	C	B	B	B	A	BC	B	D	FCM
295	179	F	169834	D	A	A	C	B	B	B	B	B	B	D	FCM
296	179	F	169835	D	A	A	C	B	B	B	A	BC	B	D	FCM
297	179	F	169836	D	A	A	C	B	B	B	AB	BC	B	D	FCM
298	179	F	169837	D	A	A	C	B	B	B	B	BC	B	D	FCM
299	179	M	169838	D	A	A	C	B	B	B	—	B	B	D	—
300)	180	F	160113	D	A	A	C	B	B	—	—	C	B	D	651
301	180	M	160114	D	A	A	C	B	B	—	—	C	B	D	632
302	180	M	160115	D	A	A	C	B	B	—	B	C	B	D	670
303	180	M	160116	D	A	A	C	B	B	—	AB	B	B	D	756
304	180	M	160117	D	A	A	C	B	B	—	—	B	B	D	774
305	180	F	160118	D	A	A	C	B	B	—	—	C	B	D	705
306	180	M	160119	D	A	A	C	B	B	—	—	C	B	D	690
307	180	M	160120	D	A	A	C	B	B	—	B	C	B	D	715
308	180	M	160121	D	A	A	C	B	B	—	AB	B	B	D	595
309	180	M	160122	D	A	A	C	B	B	—	—	B	B	D	708
310)	180	F	160123	D	A	A	C	B	B	—	AB	C	B	D	618
311	180	M	160124	D	A	A	C	B	B	—	A	C	B	D	704
312	180	M	160125	D	A	A	C	B	B	—	A	C	B	D	719
313	180	M	160126	D	A	A	C	B	B	—	AB	B	B	D	663
314	180	M	160127	D	A	A	C	B	B	—	—	B	B	D	743
315	180	F	160128	D	A	A	C	B	B	—	A	C	B	D	691
316	180	M	160129	D	A	A	C	B	B	—	A	C	B	D	705
317	180	M	160130	D	A	A	C	B	B	—	B	C	B	D	743
318	180	M	160131	D	A	A	C	B	B	—	AB	BC	B	D	635
319	180	M	160132	D	A	A	C	B	B	—	—	B	B	D	686
320)	180	F	160133	D	A	A	C	B	B	—	—	C	B	D	709
321	180	M	160134	D	A	A	C	B	B	—	—	C	B	D	740
322	180	M	160135	D	A	A	C	B	B	—	B	C	B	D	714
323	180	M	160136	D	A	A	C	B	B	—	AB	B	B	D	648
324	180	M	160137	D	A	A	C	B	B	—	—	B	B	D	729
325	181	M	160138	D	A	A	C	B	B	—	—	BC	B	D	861
326	181	M	163390	D	A	A	C	B	B	B	AB	BC	B	D	846
327	181	F	163391	D	A	A	C	B	B	B	—	C	B	D	812
328	181	M	163392	D	A	A	C	B	B	B	A	B	B	D	834
329	181	M	163393	D	A	A	C	B	B	B	—	C	B	D	786
330)	181	F	163394	D	A	A	C	B	B	B	—	C	B	D	813
331	181	M	163395	D	A	A	C	B	B	B	—	B	B	D	874
332	181	M	163396	D	A	A	C	B	B	—	AB	C	B	D	802
333	181	F	163397	D	A	A	C	B	B	B	A	C	B	D	820
334	181	M	163398	D	A	A	C	B	B	B	AB	C	B	D	885

APPENDIX 2-2
(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
335	181	F	163399	D	A	A	C	B	B	B	—	C	B	D	759
336	181	F	163400	D	A	A	C	B	B	B	A	BC	B	D	830
337	181	M	163401	D	A	A	C	B	B	B	A	BC	B	D	939
338	181	F	163402	D	A	A	C	B	B	B	—	C	B	D	836
339	181	M	163403	D	A	A	C	B	B	B	AB	B	B	D	—
340)	181	M	163404	D	A	A	C	B	B	B	A	C	B	D	702
341	182	M	163405	D	A	A	C	B	B	B	B	BC	B	D	801
342	182	M	163406	D	A	A	C	B	B	B	—	B	B	D	844
343	182	M	163407	D	A	A	C	B	B	B	B	BC	B	D	728
344	182	M	163408	D	A	A	C	B	B	B	B	B	B	D	792
345	182	M	163409	D	A	A	C	B	B	B	B	BC	B	D	806
346	182	M	163410	D	A	A	C	B	B	B	A	BC	B	D	753
347	182	F	163411	D	A	A	C	B	B	B	—	B	B	D	736
348	182	M	163412	D	A	A	C	B	B	—	B	B	B	D	782
349	182	M	163413	D	A	A	C	B	B	B	B	BC	B	D	796
350)	182	M	163414	D	A	A	C	B	B	B	B	B	B	D	805
351	182	M	163415	D	A	A	C	B	B	B	B	B	B	D	877
352	182	M	163416	D	A	A	C	B	B	B	B	BC	B	D	808
353	182	F	163417	D	A	A	C	B	B	—	B	B	B	D	791
354	182	F	163418	D	A	A	C	B	B	B	AB	BC	B	D	780
355	182	F	163419	D	A	A	C	B	B	—	AB	B	B	D	887
356	182	F	163420	D	A	A	C	B	B	—	B	BC	B	D	757
357	182	F	163421	D	A	A	C	B	B	—	B	B	B	D	766
358	182	F	163422	D	A	A	C	B	B	B	A	B	B	D	812
359	182	F	163423	D	A	A	C	B	B	B	B	BC	B	D	768
360)	182	F	163424	D	A	A	C	B	B	B	B	BD	B	D	728
361	182	F	163425	D	A	A	C	B	B	B	B	BC	B	D	811
362	182	M	163426	D	A	A	C	B	B	B	AB	BD	B	D	712
363	182	F	163427	D	A	A	C	B	B	B	—	BD	B	D	699
364	182	M	163428	D	A	A	C	B	B	B	—	C	B	D	746
365	182	M	163429	D	A	A	C	B	B	B	—	BC	B	D	754
366	182	M	163430	D	A	A	C	B	B	B	—	BC	B	D	859
367	183	F	163436	D	A	A	C	B	B	B	AB	B	B	D	714
368	183	F	163437	D	A	A	C	B	B	B	B	B	B	D	752
369	183	F	163438	D	A	A	C	B	BD	B	B	B	B	D	710
370)	183	M	163439	D	A	A	C	B	B	B	B	B	B	D	767
371	183	M	163440	D	A	A	C	B	B	B	B	B	B	D	707
372	183	F	163441	D	A	A	C	B	B	B	AB	B	B	D	802
373	183	?	163442	D	A	A	C	B	B	B	B	C	B	D	694
374	183	M	163443	D	A	A	C	B	B	B	B	BC	B	D	744
375	183	M	163444	D	A	A	C	B	B	B	AB	BD	B	D	754
376	183	?	163445	D	A	A	C	B	B	B	A	BC	B	D	760
377	183	F	163446	D	A	A	C	B	B	B	A	BD	B	D	735
378	183	M	163447	D	A	A	C	B	B	B	AB	BD	B	D	737
379	183	F	163448	D	A	A	C	B	B	B	AB	B	B	D	688
380)	183	M	163449	D	A	A	C	B	BD	B	A	BC	B	D	786
381	183	M?	163450	D	A	A	C	B	BD	B	AB	BC	B	D	910
382	183	F	163451	D	A	A	C	B	BD	B	A	C	B	D	860
383	184	M	163431	D	A	A	C	B	B	B	—	C	B	D	965
384	184	M	163432	D	A	A	C	B	B	B	AB	B	B	D	728
385	185	M	160139	D	A	A	C	B	B	—	—	BC	B	D	703
386	185	M	160140	D	A	A	C	B	B	B	B	B	B	D	689

APPENDIX 2-2
(Continued)

				Locus												
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
387	185	M	160141	D	A	A	C	B	B	B	A	C	B	D	616	
388	185	M	160142	D	A	A	C	B	B	B	A	B	B	D	686	
389	185	M	160143	D	A	A	C	B	B	B	QA	B	B	D	753	
390)	185	M	167080	D	A	A	C	B	B	B	—	B	B	D	643	
391	185	M	167081	D	A	A	C	B	B	B	—	C	B	D	758	
392	185	M	167082	D	A	A	C	B	B	B	—	C	B	D	769	
393	185	F	167083	D	A	A	C	B	B	B	—	C	B	D	770	
394	185	M	167084	D	A	A	C	B	B	B	—	BC	B	D	823	
395	185	M	167085	D	A	A	C	B	B	B	—	B	B	D	1063	
396	185	M	167086	D	A	A	C	B	B	B	—	C	B	D	765	
397	185	M	167087	D	A	A	C	B	B	B	—	C	B	D	671	
398	185	M	167088	D	A	A	C	B	B	B	—	BC	B	D	751	
399	185	M	167089	D	A	A	C	B	B	B	—	BC	B	D	736	
400)	185	F	167090	D	A	A	C	B	B	B	B	BC	B	D	760	
401	185	F	167091	D	A	A	C	B	B	B	—	BC	B	D	842	
402	185	F	167092	D	A	A	C	B	B	B	A	C	B	D	725	
403	186	M	160144	D	A	A	C	B	B	B	AB	B	B	D	672	
404	186	F	160145	D	A	A	C	B	B	—	A	—	—	D	611	
405	186	M	160146	D	A	A	C	B	B	B	AB	B	B	D	624	
406	186	M	160147	D	A	A	C	B	B	B	AB	B	B	D	697	
407	186	M	160148	D	A	A	C	B	B	B	A	BC	B	D	658	
408	186	M	160149	D	A	A	C	B	B	B	A	B	B	D	614	
409	186	M	160150	D	A	A	C	B	B	B	—	C	B	D	633	
410)	186	M	160151	D	A	A	C	B	B	—	AC	B	B	D	664	
411	186	M	160152	D	A	A	C	B	B	—	A	BC	B	D	708	
412	186	F	160153	D	A	A	C	B	B	B	A	B	B	D	620	
413	186	M	160154	D	A	A	C	B	B	B	BC	B	B	D	630	
414	186	M	160155	D	A	A	C	B	B	—	A	BC	B	D	650	
415	186	M	160156	D	A	A	C	B	B	—	A	B	B	D	595	
416	186	M	160157	D	A	A	C	B	B	—	—	BC	B	D	690	
417	186	F	167093	D	A	A	C	B	B	B	—	BC	B	D	722	
418	186	F	167094	D	A	A	C	B	B	B	—	B	B	D	741	
419	186	F	167095	D	A	A	C	B	B	B	—	B	B	D	693	
420)	186	F	167096	D	A	A	C	B	B	B	—	BC	B	D	1031	
431	187	F	163452	D	A	A	C	B	B	B	A	B	B	D	784	
422	187	F	163453	D	A	A	C	B	B	B	A	B	B	D	783	
423	187	F	163454	D	A	A	C	B	B	B	A	B	B	D	731	
424	187	F	163455	D	A	A	C	B	B	B	A	B	B	D	750	
425	187	M	163456	D	A	A	C	B	B	B	A	B	B	D	732	
426	187	M	167073	D	A	A	C	B	B	B	—	B	B	D	778	
427	187	F	167074	D	A	A	C	B	B	B	—	C	B	D	643	
428	187	M	167075	D	A	A	C	B	BD	B	—	B	B	D	821	
429	187	F	167076	D	A	A	C	B	B	B	—	B	B	D	768	
430)	187	F	167077	D	A	A	C	B	B	B	—	B	B	D	771	
431	187	F	167078	D	A	A	C	B	B	B	—	B	B	D	763	
432	187	M	167079	D	A	A	C	B	B	B	—	B	B	D	770	
433	188	M	165856	D	A	A	C	B	B	—	AC	BC	B	D	876	
434	188	M	165857	D	A	A	C	B	B	—	AB	—	B	D	749	
435	188	M	165858	D	A	A	C	B	B	—	AC	BC	B	D	875	
436	188	M	165859	D	A	A	C	B	B	—	AB	BC	B	D	650	
437	188	M	165860	D	A	A	C	B	B	—	AB	B	B	D	635	
438	188	M	165861	D	A	A	C	B	B	—	AC	BC	B	D	694	

APPENDIX 2-2
(Continued)

Locus														
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
439	188	M	165862	D	A	A	C	B	B	—	A	—	B	D 681
440)	188	M	165863	D	A	A	C	B	B	—	AB	BC	B	D 758
441	188	M	165864	D	A	A	C	B	B	—	A	BC	B	D 771
442	188	M	165865	D	A	A	C	B	B	B	—	C	B	D 708
443	190	M	JPB25772	D	A	A	C	B	B	—	AB	C	B	D —
444	190	M	JPB25773	D	A	A	C	B	B	—	A	B	B	D 770
445	190	M	JPB25774	D	A	A	C	B	B	—	AB	B	B	D 648
446	190	M	JPB25775	D	A	A	C	B	B	—	A	BC	B	D 826
447	191	M	163457	D	A	A	C	B	B	—	A	BC	B	D 831
448	191	M	163459	D	A	A	C	B	B	B	B	BC	B	D 880
449	191	M	163460	D	A	A	C	B	B	B	A	B	B	D 838
450)	192	M	160158	D	A	A	C	B	B	B	—	BC	B	D 904
451	194	M	165866	D	A	A	C	B	B	—	A	B	B	D 711
452	195	M	158779	D	A	A	C	B	B	B	—	B	B	D 808
453	195	M	158780	D	A	A	C	B	B	B	—	B	B	D 763
454	195	F	158781	D	A	A	C	B	B	B	—	B	B	D 724
455	195	F	158782	D	A	A	C	B	B	B	—	B	B	D 764
456	195	M	158783	D	A	A	C	B	B	B	—	B	B	D 757
457	195	M	158784	D	A	A	C	B	B	B	—	B	B	D 783
458	196	M	160159	D	A	A	C	B	B	B	—	B	B	D 785
459	196	F	160160	D	A	A	C	B	B	B	AB	B	B	D 721
460)	196	F	160161	D	A	A	C	B	B	B	—	B	B	D 743
461	203	M	165867	D	A	A	C	B	B	—	A	B	B	D 746
462	205	F	163373	D	A	QA	C	B	B	B	A	—	B	D 826
463	212	M	163374	D	A	A	C	B	B	BC	A	B	B	D 728
464	212	F	163375	D	A	A	C	B	B	B	B	—	—	D 732

Appendix 2-3: Genotypes of diploid (2n) *Ambystoma laterale-jeffersonianum* LJ unisexual specimens ordered by site number.

Locus														
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
1	18	F	165937	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 765
2	107	F	169913	BD	AB	AB	BC	A	BD	—	BC	B	B	BD FCM
3	107	F	169914	BD	AB	AB	BC	B	BD	—	BC	B	B	BD FCM
4	107	F	169915	BD	AB	AB	BC	A	BD	—	BC	BC	B	BD FCM
5	107	F	169916	BD	AB	AB	BC	A	BD	—	AC	B	B	BD FCM
6	108	F	160248	BD	AB	AB	BC	B	BD	AB	AC	B	B	BD 752
7	108	F	160249	BD	AB	AB	BC	B	BD	AB	A	BC	B	BD 836
8	108	F	160250	BD	AB	AB	BC	B	BD	—	AC	B	B	BD 768
9	108	F	160251	BD	AB	AB	BC	B	BD	AB	AC	B	B	BD 860
10)	108	F	160252	BD	AB	AB	BC	B	BD	—	AC	BC	B	BD 755
11	108	F	160253	BD	AB	AB	BC	A	BD	—	AC	B	B	BD 760
12	108	F	160254	BD	AB	AB	BC	B	BD	—	AC	B	B	BD 707
13	109	F	169917	BD	AB	AB	BC	B	BD	—	AC	B	B	BD FCM
14	109	F	169918	BD	AB	AB	BC	B	BD	—	AC	B	B	BD FCM
15	110	F	169395	BD	AB	AB	BC	B	BD	AB	BC	—	B	BD FCM
16	112	F	169802	BD	AB	B	BC	B	BD	BD	AC	B	B	BD FCM

APPENDIX 2-3
(Continued)

Locus														
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
17	113	F	162819	BD	AB	AB	BC	B	BD	BC	BC	B	B	BD 787
18	113	F	162820	BD	AB	AB	BC	B	BD	BC	AC	B	B	BD 696
19	113	F	162821	BD	AB	AB	BC	B	BD	BC	C	BC	B	BD 726
20)	113	F	162823	BD	AB	AB	BC	B	BD	BC	C	BC	B	BD 782
21	113	F	162824	BD	AB	AB	BC	B	BD	BC	C	BC	B	BD 860
22	113	F	162825	BD	AB	AB	BC	B	BD	—	AC	B	B	BD —
23	113	F	162826	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 893
24	113	F	162827	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 760
25	113	F	162828	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 789
26	113	F	162829	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 797
27	113	F	162830	BD	AB	AB	BC	B	BD	BC	AC	B	B	BD 868
28	113	F	162862	BD	AB	AB	BC	B	BD	—	—	BC	B	BD 802
29	113	F	164590	BD	AB	AB	BC	B	BD	—	—	BC	B	D 759
30)	113	F	164591	BD	AB	AB	BC	B	BD	BC	C	BC	B	BD 718
31	113	F	164592	BD	AB	AB	BC	B	BD	BC	AC	B	B	BD 642
32	113	F	164636	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD 978
33	113	F	164637	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD 758
34	113	F	164638	BD	AB	AB	BC	B	BD	BC	C	BC	B	BD 836
35	114	M	153110	BD	AB	A	C	B	BD	BC	—	B	D	BD 708
36	122	F	153107	BD	AB	AB	BC	—	BD	B	—	C	B	D 766
37	122	M	153108	BD	AB	AB	BC	B	BD	—	BC	BC	B	BD 736
38	122	F	153109	BD	AB	AB	BC	B	BD	—	—	B	B	BD 740
39	122	F	153119	BD	AB	AB	BC	—	BD	AB	—	B	B	BD 823
40)	123	F	153111	BD	AB	AB	BC	—	BD	—	—	BC	B	BD 702
41	123	F	162832	BD	AB	AB	BC	B	BD	—	—	B	B	BD 689
42	123	F	162833	BD	AB	AB	B	B	BD	—	—	B	B	BD 711
43	123	F	162834	BD	AB	AB	BC	B	BD	—	—	B	B	BD 690
44	123	F	165938	BD	AB	AB	BC	B	BD	—	—	B	B	BD —
45	123	F	165939	BD	AB	AB	BC	B	BD	—	—	B	B	BD 612
46	123	F	165940	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 572
47	123	F	165941	BD	AB	AB	BC	B	BD	—	—	B	B	BD 615
48	123	F	165942	BD	AB	AB	B	B	BD	BC	—	B	B	BD 647
49	123	F	165943	BD	AB	AB	BC	B	BD	—	—	B	B	BD 602
50)	123	F	165944	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 649
51	124	F	169810	BD	AB	AB	BC	B	BD	AB	—	B	B	BD FCM
52	124	F	169811	BD	AB	AB	BC	AB	BD	AB	AC	BC	B	BD FCM
53	124	F	169812	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD FCM
54	125	F	169813	BD	AB	AB	BC	AB	BD	BC	AC	BC	B	BD FCM
55	125	F	169814	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD FCM
56	126	F	169772	BD	AB	AB	BC	B	BD	BC	—	BC	B	BD FCM
57	126	F	169773	BD	AB	AB	BC	B	BD	BC	—	BC	B	BD FCM
58	126	F	169774	BD	AB	AB	BC	B	BD	BC	—	BC	B	BD FCM
59	126	F	169775	BD	AB	AB	BC	B	BD	BD	—	BC	B	BD FCM
60)	126	F	169776	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD FCM
61	130	F	165945	BD	AB	QB	BC	B	BD	BC	—	BC	B	BD 816
62	134	F	162831	BD	AB	AB	BC	B	BD	—	—	B	B	BD 917
63	137	F	167065	BD	AB	AB	BC	B	BD	BC	—	B	B	BD 661
64	137	F	167066	BD	AB	AB	—	B	—	BC	AC	B	B	BD —
65	138	F	167070	BD	AB	AB	BC	B	—	BC	—	B	B	BD 771
66	156	F	162816	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD 778
67	156	F	162817	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD 778
68	156	F	162818	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD 670

APPENDIX 2-3
(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
69	158	F	165946	BD	AB	A	BC	B	BD	BC	—	BC	B	BD	561
70)	159	F	160225	BD	AB	AB	BC	B	BD	BC	AB	BC	B	D	740
71	169	F	167006	BD	AB	AB	BC	B	BD	BC	—	BC	B	BD	752
72	174	F	169928	BD	AB	AB	BC	B	BD	—	AC	B	B	BD	FCM
73	192	F	169767	BD	AB	AB	BC	B	BD	AB	—	B	B	BD	FCM
74	201	F	165947	BD	AB	AB	BC	B	BD	—	—	B	B	BD	660
75	201	F	165948	BD	AB	AB	BC	C	BD	BC	—	B	B	BD	700
76	201	F	165949	BD	AB	AB	BC	C	BD	—	—	B	B	BD	789
77	201	F	165950	BD	AB	AB	BC	C	BD	BC	—	B	B	BD	762
78	201	F	165951	BD	AB	AB	BC	C	BD	—	—	B	B	BD	667
79	201	F	165952	BD	AB	AB	BC	C	BD	BC	—	B	B	BD	574
80)	201	F	169766	BD	AB	AB	BC	BC	BD	BC	—	B	B	BD	FCM
81	211	F	160246	BD	AB	AB	BC	B	BD	BC	—	B	B	BD	742
82	211	F	160247	BD	AB	AB	BC	B	BD	BC	—	B	B	BD	696
83	214	F	158785	BD	AB	AB	BC	B	BD	BC	AC	BC	B	BD	729
84	214	F	158786	BD	AB	AB	BC	B	BD	BC	—	BC	B	BD	732
85	215	F	158812	BD	AB	AB	BC	B	BD	AB	AC	B	B	BD	611
86	215	F	158816	BD	AB	AB	BC	B	BD	AB	AC	B	B	BD	638

Appendix 2-4: Genotypes of triploid (3n) *Ambystoma 2 laterale* – *jeffersonianum* (LLJ) specimens ordered by site number.

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
1	7	F	164593	BBD	ABB	ABB	BBC	ABB	BDD	BCC	—	B	B	BBD	983
2	18	F	165994	BBD	ABB	ABB	BBC	B	BDD	—	—	—	—	—	951
3	42	F	166983	BBD	AAB	ABB	BBC	B	BBD	BCC	—	BBC	B	BBD	990
4	42	F	166984	BBD	ABB	ABB	BBC	B	BBD	BCC	—	B	B	BBD	964
5	42	F	166985	BBD	ABB	ABB	BBC	B	BBD	BCC	—	B	B	BBD	1093
6	42	F	166986	BBD	ABB	ABB	BBC	ABB	BDD	BCC	BCC	B	B	BBD	1006
7	42	F	166987	BBD	ABB	ABB	BBC	B	BBD	BCC	—	BBC	B	BBD	902
8	107	F	169900	BBD	ABB	ABB	BBC	A	BDD	—	BCC	B	B	BBD	FCM
9	107	F	169901	BBD	ABB	B	BBC	A	BDD	—	BCC	BBC	B	BBD	FCM
10)	107	F	169902	BBD	ABB	ABB	BBC	B	BDD	—	BCC	B	B	BBD	FCM
11	107	F	169903	BBD	ABB	B	BBC	B	BDD	—	BCC	B	B	BBD	FCM
12	107	F	169904	BBD	ABB	ABB	BBC	B	BDD	—	BCC	B	B	BBD	FCM
13	107	F	169905	BBD	ABB	ABB	BBC	A	BDD	—	BCC	B	B	BBD	FCM
14	107	F	169906	BBD	ABB	ABB	BBC	B	BDD	—	ABC	B	B	BBD	FCM
15	108	M	160055	BBD	—	B	BBC	B	BDD	—	—	B	B	BBD	796
16	108	F	160269	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	BBC	B	BBD	1058
17	108	F	160270	BBD	ABB	ABB	B	B	BDD	BCC	—	BBC	B	BBD	996
18	108	F	160271	BBD	ABB	B	BBC	B	BDD	BCC	—	B	B	BBD	1178
19	108	F	160272	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1195
20)	108	F	160273	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	1170
21	108	F	160274	BBD	ABB	ABB	BBC	A	BDD	BCC	AAC	B	B	BBD	1194
22	108	F	160275	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	BCC	B	BBD	1061

APPENDIX 2-4
(Continued)

	Site	Sex	AMNH	Locus												blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1		
23	108	F	160276	BBD	ABB	B	BBC	B	BDD	BCC	ACC	BBC	B	BBD	792	
24	108	F	160277	BBD	ABB	ABB	BBC	B	BDD	—	AAC	B	B	BBD	1137	
25	108	F	160278	BBD	ABB	ABB	BBC	A	BDD	BCC	AAC	BBC	B	BBD	1029	
26	108	F	160279	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	BCC	B	BBD	1062	
27	108	F	160280	BDD	ABB	ABB	BBC	B	BDD	BCC	AAC	BBC	B	BBD	950	
28	108	F	160281	BDD	ABB	ABB	BBC	A	BDD	BCC	AAC	BCC	B	BBD	893	
29	108	F	160282	BDD	ABB	ABB	BBC	A	BBD	BCC	AAC	BBC	B	BBD	913	
30)	108	F	160283	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BCC	B	BBD	1186	
31	108	F	160284	BBD	ABB	ABB	BBC	A	BDD	—	AAC	B	B	BBD	1024	
32	110	F	165995	BBD	ABB	ABB	BBC	A	BDD	BCC	—	B	B	BBD	959	
33	110	F	169396	BBD	ABB	—	—	—	BDD	A	—	B	B	BBD	FCM	
34	110	F	169397	BBD	ABB	ABB	BBC	B	BDD	BCC	C	B	B	BBD	FCM	
35	110	F	169398	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	FCM	
36	110	F	169894	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	BBC	B	BBD	FCM	
37	110	F	169895	BBD	ABB	AAB	BBC	B	BBD	BCC	ACC	BBC	B	BBD	FCM	
38	110	F	169896	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	BBC	B	BBD	FCM	
39	110	F	169897	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	FCM	
40)	110	F	169898	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	FCM	
41	111	F	169385	BBD	ABB	ABB	BBC	B	BDD	C	BCC	B	B	B	FCM	
42	111	F	169399	BBD	ABB	ABB	BBC	BBC	BBD	BCC	ACC	B	B	BBD	FCM	
43	111	F	169400	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	FCM	
44	111	F	169401	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	FCM	
45	111	F	169402	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	FCM	
46	111	F	169403	BBD	ABB	—	BBC	B	BDD	ABB	—	B	B	BBD	FCM	
47	111	F	169404	BBD	ABB	—	—	—	BDD	BCC	—	B	B	BBD	FCM	
48	111	F	169405	BBD	ABB	—	—	—	BDD	BCC	—	B	B	BBD	FCM	
49	111	F	169406	BBD	ABB	—	BBC	B	BDD	BCC	—	B	B	BBD	FCM	
50)	111	F	169407	BBD	ABB	—	BBC	B	BDD	BCC	—	B	B	BBD	FCM	
51	111	F	169408	BBD	ABB	ABB	BBC	B	BDD	B	ACC	B	B	B	FCM	
52	111	F	169409	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	B	B	BBD	FCM	
53	111	F	169410	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	FCM	
54	111	F	169806	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	FCM	
55	112	F	169803	BBD	ABB	B	BBC	B	BD	BCC	ACC	B	B	BBD	FCM	
56	112	F	169804	BBD	ABB	B	BBC	B	BDD	BCC	—	B	B	BBD	FCM	
57	112	F	169805	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	FCM	
58	112	M	169807	BBD	ABB	B	B	B	BDD	BCC	—	BBC	B	BBD	—	
59	113	F	162942	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	BBC	B	BBD	994	
60)	113	F	162943	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	1043	
61	113	F	162944	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	B	B	BBD	994	
62	113	F	162945	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	B	B	BBD	1027	
63	113	F	162946	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BCC	B	BBD	806	
64	113	F	162947	BBD	ABB	AAB	BBC	B	BDD	BCC	—	B	B	BBD	1196	
65	113	F	162948	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBD	B	BBD	1154	
66	113	F	162949	BBD	ABB	ABB	AAC	B	BDD	BCC	ACC	B	B	BBD	923	
67	113	F	162951	BBD	ABB	A	BBC	B	BBD	BCC	ACC	B	B	BBD	1005	
68	113	F	162952	BBD	ABB	AAB	BBC	B	BDD	BBC	AAC	B	B	BBD	982	
69	113	F	162953	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	1038	
70)	113	F	162954	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1110	
71	113	F	162955	BDD	ABB	A	BBC	B	BBD	BCC	ACC	B	B	BBD	833	
72	113	F	162956	BBD	ABB	A	BBC	B	BBD	—	BCC	B	B	BBD	906	
73	113	F	162957	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	B	B	BBD	996	
74	113	F	162958	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	962	

APPENDIX 2-4

(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
75	113	F	162959	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	925
76	123	F	165996	BBD	ABB	ABB	BBC	B	BBD	—	—	—	—	—	670
77	125	F	169815	BBD	ABB	ABB	BBC	ABB	D	BCC	BCC	BBC	B	BBD	FCM
78	127	F	160257	BBD	ABB	ABB	BCC	BBC	BDD	BCC	BCC	B	B	BBD	1056
79)	127	F	160258	BBD	ABB	ABB	BCC	BBC	BDD	C	AAC	BBC	B	BBD	929
80	127	F	160259	BBD	ABB	ABB	BBC	B	BDD	C	BCC	BBC	B	BBD	1199
82	127	F	160260	BBD	ABB	ABB	B	B	BDD	—	—	BBC	B	BBD	1007
83	127	F	160261	BBD	ABB	ABB	BBC	B	BDD	—	—	B	B	BBD	1252
84	127	F	160262	BBD	ABB	ABB	—	B	BDD	—	—	BBC	B	BBD	1032
81	127	F	160263	BBD	ABB	ABB	BBC	B	BDD	C	C	BBC	B	B	1121
85	127	F	160264	BBD	ABB	ABB	BBC	B	BDD	C	BCC	BBC	B	BBD	1048
86	127	F	160265	BBD	ABB	ABB	BBC	B	BDD	—	—	B	B	BBD	926
87	127	F	160266	BBD	ABB	ABB	—	—	BDD	—	—	BBC	B	BBD	988
88	128	F	160268	BBD	ABB	ABB	BBC	A	BDD	—	—	B	B	BBD	911
89	129	F	162961	BBD	ABB	ABB	BBC	B	BDD	—	C	B	B	BBD	1235
90)	129	F	165997	BBD	ABB	ABB	BCC	B	BDD	C	C	BBC	B	BBD	996
91	129	F	165998	BBD	ABB	ABB	BBC	B	BDD	C	C	B	B	BBD	916
92	129	F	165999	BBD	ABB	ABB	BBC	B	BDD	C	C	BBC	B	BBD	1063
93	132	F	162960	BBD	ABB	ABB	BBC	B	BDD	C	AAC	BBC	B	BBD	909
94	133	F	166000	BBD	ABB	ABB	BCC	B	BDD	BCC	C	BBC	B	BBD	841
95	133	F	166001	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	828
96	133	F	166002	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1088
97	133	F	166003	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	1184
98	136	F	166004	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	944
99	136	F	166005	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	966
100)	136	F	166006	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	987
101	136	F	166007	BBD	ABB	B	BBC	B	BDD	BCC	—	BBC	B	BBD	1071
102	136	F	166008	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1031
103	136	F	166009	BBD	ABB	ABB	BBC	B	BDD	—	C	B	B	BBD	922
104	136	F	166010	BBD	ABB	ABB	BBC	B	BDD	—	C	BBC	B	BBD	1001
105	136	F	166011	BBD	ABB	ABB	BBC	B	BDD	—	C	BBC	B	BBD	1000
106	136	F	166012	BBD	ABB	ABB	BBC	B	BDD	—	ACC	BBC	B	BBD	1200
107	136	F	166013	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	879
108	136	F	166014	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1033
109	143	F	167033	BBD	ABB	—	—	—	—	BCC	—	B	B	BBD	1194
110)	143	F	167034	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	1022
111	143	F	167035	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	1018
112	143	F	167036	BBD	ABB	—	BBC	B	BDD	BCC	BCC	B	B	BBD	1065
113	144	F	166015	BBD	ABB	AAB	BBC	B	BDD	—	C	B	B	BBD	689
114	144	F	166016	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	951
115	144	F	166017	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	971
116	145	F	167041	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	990
117	145	F	167042	BBD	ABB	ABB	BCC	B	BDD	BCC	BCC	B	B	BBD	997
118	145	F	167043	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	—	—	BBD	1046
119	145	F	167044	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	1069
120)	145	F	167045	BBD	ABB	ABB	BCC	B	BDD	BCC	ABC	—	—	BBD	1038
121	145	F	167046	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	985
122	145	F	167047	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	BBC	B	BBD	967
123	145	F	167048	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	1110
124	145	F	167049	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	B	B	BBD	FCM
125	145	F	167050	BBD	ABB	ABB	BBC	B	BDD	BCC	ABC	BCC	B	BBD	878
126	145	F	167051	BBD	ABB	B	BBC	B	BDD	BCC	ABC	BCC	B	BBD	913

APPENDIX 2-4
(Continued)

Locus																
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
127	145	F	167052	BBD	ABB	ABB	BBC	B	BDD	—	ABC	B	B	BBD	1175	
128	145	F	167053	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	B	B	BBD	909	
129	145	F	167054	BBD	ABB	ABB	BBC	B	BDD	—	ABC	B	B	BBD	1120	
130)	145	F	167055	BBC	ABB	ABB	BBC	B	BDD	—	ACC	B	B	BBD	985	
131	148	F	167012	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BDD	1041	
132	148	F	167013	BBD	ABB	ABB	BBC	B	BDD	BCC	BBC	—	—	BBD	984	
133	148	F	167014	BBD	ABB	ABB	BBC	B	BDD	C	BCC	B	B	BBD	1021	
134	148	F	167015	BBD	ABB	ABB	D	B	BDD	BCC	BBC	—	—	BBD	977	
135	148	F	167016	BBD	ABB	B	BBC	B	D	BCC	ABC	ABC	B	BBD	1180	
136	148	F	167017	BBD	ABB	ABB	D	AAB	BDD	BCC	BCC	—	—	BBD	944	
137	148	F	167018	BBD	ABB	B	BBC	B	BDD	BCC	BCC	—	—	BBD	1126	
138	148	F	167019	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	1164	
139	149	F	169446	BBD	ABB	ABB	BBC	B	BBD	BCC	AAC	BBC	B	BBD	FCM	
140)	151	F	169447	BBD	ABB	ABB	BBC	B	BDD	BCC	BBC	BBC	B	BBD	FCM	
141	151	F	169448	BBD	ABB	ABB	BBC	B	BDD	BCC	AAC	BBC	B	BBD	FCM	
142	152	M	169449	BBD	ABB	—	—	B	BDD	BCC	—	BBC	B	BBD	FCM	
143	152	F	169450	BBD	ABB	—	BBC	ABB	BDD	BBC	—	BBC	B	BBD	FCM	
144	155	F	160285	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1057	
145	155	F	160286	BBD	ABB	ABB	BBC	ABB	BDD	BCC	ACC	BBC	B	BBD	1022	
146	155	F	160287	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1094	
147	155	F	160288	BBD	ABB	ABB	BBC	B	BDD	—	ACC	BBC	B	BBD	1013	
148	155	F	160289	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BBC	B	BBD	951	
149	155	F	160290	BBD	ABB	B	C	B	BDD	BCC	QCC	—	B	BBD	985	
150)	155	F	160291	BBD	ABB	ABB	BBC	B	BDD	—	ACC	BBC	B	BBD	876	
151	161	F	166999	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	1043	
152	161	F	167000	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	1067	
153	161	F	167001	—	—	—	—	—	BDD	BCC	—	B	B	BBD	946	
154	162	F	167002	BBD	ABB	ABB	BBC	ABB	BDD	BCC	—	—	—	BBD	1078	
155	162	F	167003	BBD	ABB	ABB	BBC	B	BDD	BCC	—	—	—	BBD	928	
156	162	F	169359	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BBC	B	BBD	FCM	
157	162	F	169360	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BC	B	BBD	FCM	
158	162	F	169361	BBD	ABB	ABB	BBC	B	BDD	BCC	BCC	B	B	BBD	FCM	
159	162	M	169362	BBD	ABB	ABB	BBC	AAB	BDD	BCC	AAC	BBC	B	BBD	FCM	
160)	162	F	169363	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BBC	B	BBD	FCM	
161	162	F	169364	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	FCM	
162	162	F	169365	BBD	ABB	ABB	BBC	B	BDD	BCC	A	B	B	BBD	FCM	
163	162	F	169366	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	BBC	B	BBD	FCM	
164	168	F	160292	BBD	ABB	ABB	BBC	A	BDD	—	AAC	BBC	B	BBD	1071	
165	168	F	160293	BBD	ABB	ABB	BBC	B	BDD	—	—	BBC	B	BBD	1012	
166	168	F	160294	BBD	ABB	ABB	BBC	A	BDD	—	AAC	BBC	B	BBD	1099	
167	168	F	160295	BBD	ABB	ABB	BBC	A	BDD	—	ACC	BBC	B	BBD	1047	
168	168	F	160296	BBD	ABB	B	BBC	B	BDD	—	—	BBC	B	BBD	962	
169	168	F	160297	BBD	ABB	ABB	BBC	B	BDD	—	—	BBC	B	BBD	946	
170)	168	F	160298	BBD	ABB	ABB	BBC	B	BDD	—	—	BBC	B	BBD	982	
171	168	F	160299	BBD	—	ABB	BBC	B	BDD	—	—	BBC	B	BBD	966	
172	168	F	160300	BBD	—	ABB	BBC	B	BDD	—	—	BBC	B	BBD	905	
173	168	F	160301	BBD	—	B	BBC	B	BDD	—	—	BBC	B	BBD	803	
174	168	F	160302	BBD	ABB	ABB	BBC	AAB	BDD	BCC	—	BBC	B	BBD	946	
175	168	F	160303	BBD	ABB	ABB	BBC	AAB	BDD	—	AAC	BBC	B	BBD	1062	
176	168	F	160304	BBD	ABB	ABB	BBC	AAB	BDD	BCC	—	BBC	B	BBD	955	
177	168	F	160305	BBD	ABB	ABB	BBC	ABB	BDD	BCC	—	BBC	B	BBD	911	
178	168	F	160306	BBD	ABB	ABB	BBC	ABB	BDD	BCC	—	BBC	B	BBD	1007	

APPENDIX 2-4
(Continued)

			Locus												
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
179	168	F	160307	BBD	ABB	B	B	B	BDD	BCC	—	BBC	B	BBD	1058
180)	168	F	160308	BBD	ABB	ABB	BBC	AAB	BDD	BCC	—	BBC	B	BBD	1072
181	168	F	160309	BBD	ABB	ABB	BBC	ABB	BDD	BCC	—	BBC	B	BBD	1070
182	168	F	160311	BBD	ABB	ABB	BBC	AAB	BDD	—	ACC	BBC	B	BBD	906
183	168	F	160312	BBD	ABB	ABB	BBC	ABB	BDD	—	ACC	BBC	B	BBD	858
184	168	F	160313	BBD	ABB	ABB	BBC	AAB	BDD	BCC	—	BBC	B	BBD	931
185	168	F	160314	BBD	ABB	ABB	BBC	AAB	BDD	BCC	ACC	BBC	B	BBD	924
186	168	F	160315	BBD	ABB	ABB	BBC	ABB	BDD	BCC	ACC	BBC	B	BBD	976
187	168	F	160316	BBD	ABB	ABB	BBC	B	BDD	—	ACC	BBC	B	BBD	898
188	168	F	160317	BBD	ABB	ABB	BBC	B	BDD	—	ACC	BBC	B	BBD	937
189	168	F	160320	BBD	ABB	ABB	BBC	AAB	BDD	BCC	—	BBC	B	BBD	1017
190)	174	F	169926	BBD	ABB	ABB	BBC	B	BDD	—	ACC	B	B	BBD	FCM
191	174	F	169927	BBD	ABB	ABB	BBC	B	BDD	—	AAC	B	B	BBD	FCM
192	189	F	166018	BBD	ABB	ABB	BBC	B	BDD	BCC	C	B	B	BBD	997
193	189	F	166019	BBD	ABB	ABB	BBC	ABB	BDD	BCC	BCC	BBC	B	BBD	986
194	189	F	166020	BBD	ABB	ABB	D	B	BDD	BCC	AAC	BBC	B	BBD	1032
195	189	F	166021	BBD	ABB	ABB	BBC	ABB	BDD	BCC	BCC	BBC	B	BBD	958
196	189	F	166022	BBD	ABB	ABB	BBC	B	BDD	BCC	C	B	B	BBD	969
197	189	F	166023	BBD	ABB	ABB	BBC	B	BDD	BCC	C	B	B	BBD	1004
198	189	F	166024	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	885
199	189	F	166025	BBD	ABB	ABB	D	B	BDD	BCC	C	BBC	B	BBD	894
200)	189	F	166026	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	981
201	189	F	166027	BBD	ABB	ABB	D	B	BDD	BCC	BCC	B	B	BBD	817
202	189	F	166028	BBD	ABB	ABB	BBC	ABB	BDD	BCC	AAC	BBC	B	BBD	899
203	189	F	166029	BBD	ABB	ABB	D	ABB	BDD	BCC	ACC	B	B	BBD	842
204	189	F	166030	BBD	ABB	ABB	D	B	BDD	BCC	C	BBC	B	BBD	916
205	189	F	166031	BBD	ABB	ABB	BBC	B	BDD	BCC	C	BBC	B	BBD	999
206	189	F	166032	BBD	ABB	ABB	BBC	B	BDD	C	C	BBC	B	BBD	929
207	189	F	166033	BBD	ABB	ABB	D	B	BDD	BBC	ACC	B	B	BBD	958
208	189	F	166034	BBD	ABB	ABB	D	B	BDD	BCC	C	BBC	B	BBD	—
209	189	F	166035	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	925
210)	189	F	166036	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1198
211	189	F	166037	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	814
212	197	F	169343	BBD	ABB	ABB	BBC	B	BDD	BCC	—	B	B	BBD	FCM
213	198	F	169344	BBD	ABB	ABB	BBC	BBC	BBD	BCC	AAC	B	B	BBD	FCM
214	198	F	169345	BBD	ABB	ABB	BBC	A	BBD	BCC	AAC	B	B	BBD	FCM
215	209	F	160267	BDD	ABB	ABB	BBC	QAA	BBD	BCC	AAC	B	B	BBD	862
216	211	F	158824	BBD	ABB	ABB	BBC	B	BDD	—	—	B	B	BBD	1049
217	211	F	160256	BBD	ABB	ABB	BBC	A	BDD	BCC	—	B	B	BBD	1011
218	214	F	158811	BBD	ABB	ABB	BBC	B	BDD	BCC	—	BBC	B	BBD	1007
219	215	F	158813	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	956
220)	215	F	158814	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	—	B	BBD	960
221	215	F	158815	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1043
222	215	F	158817	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1234
223	215	F	158818	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1076
224	215	F	158819	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1027
225	215	F	158820	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	935
226	215	F	158821	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	987
227	215	F	158822	BBD	ABB	ABB	BBC	B	BDD	BCC	ACC	B	B	BBD	1179
228	216	F	153120	BBD	ABB	ABB	BCC	B	BDD	—	—	B	B	BBD	1048

Appendix 2-5: Genotypes of triploid (3n) *Ambystoma laterale* – (2) *jeffersonianum* (LJJ) specimens ordered by site number.

Locus															
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
1	20	F	169808	BDD	AAB	AAB	BCC	B	BBD	AAB	—	BBC	B	BDD	FCM
2	20	F	169809	BDD	AAB	AAB	—	B	BBD	AAB	—	BBC	B	BDD	FCM
3	28	F	158787	BDD	ABB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	982
4	108	F	153118	BDD	AAB	AAB	BCC	ABB	BBD	—	—	BCC	B	BDD	1144
5	108	F	160330	BDD	AAB	AAB	BCC	A	BBD	—	ABC	BCC	B	BDD	1009
6	109	F	169849	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BCC	B	BDD	FCM
7	109	F	169919	BDD	AAB	ABB	BCC	B	BBD	—	BBC	BCC	B	BDD	FCM
8	109	F	169920	BDD	AAB	ABB	BCC	B	BBD	—	BBC	BCC	B	BDD	FCM
9	109	F	169921	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BCC	B	BDD	FCM
10)	113	F	162939	BDD	AAB	ABB	BCC	B	BBD	BBC	—	B	B	BDD	917
11	114	F	153112	BDD	AB	ABB	BCC	B	BBD	—	—	BBC	B	BDD	—
12	115	F	153116	BDD	AAB	AAB	BCC	B	BBD	—	AAB	BBC	B	BDD	948
13	115	F	160337	BDD	AAB	AAB	BCC	A	BBD	AAB	AAC	BBC	B	BDD	1090
14	117	F	169451	BDD	AAB	A	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
15	117	F	169452	BDD	AAB	A	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
16	117	F	169453	BDD	AAB	ABB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	FCM
17	117	F	169454	BDD	AAB	A	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
18	117	F	169455	BDD	AAB	A	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
19	119	F	160331	BDD	AAB	AAB	BCC	ABB	BBD	BBC	—	BCC	B	BDD	1239
20)	119	F	160332	BDD	AAB	AAB	BCC	ABB	BBD	BBC	—	BCC	B	BDD	1148
21	119	F	160333	BDD	AAB	AAB	BCC	ABB	BBD	BBC	—	BCC	B	BDD	1503
22	119	F	160334	BDD	AAB	AAB	BCC	ABB	BBD	—	ABC	BCC	B	BDD	1321
23	119	F	160335	BDD	AAB	AAB	BCC	ABB	BBD	BBC	—	BCC	B	BDD	1358
24	119	F	160336	BDD	AAB	AAB	BCC	ABB	BBD	—	ABC	BBC	B	BDD	1283
25	122	F	153113	BDD	AAB	ABB	BCC	B	BBD	—	ABC	B	B	BDD	996
26	122	F	153114	BDD	AAB	AAB	BCC	B	BBD	—	—	BBC	B	BDD	937
27	122	F	153115	BDD	AAB	AAB	BCC	B	BBD	—	—	BBC	B	BDD	924
28	123	F	162835	BDD	AAB	AAB	BCC	B	BBD	—	—	B	B	BDD	713
29	124	F	169816	BDD	AAB	AAB	BCC	B	BBD	AAB	—	BBC	B	BDD	FCM
30)	124	F	169817	BDD	AAB	AAB	BCC	B	BBD	AAB	—	B	B	BDD	FCM
31	124	F	169818	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
32	124	F	169819	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	B	B	BDD	FCM
33	124	F	169820	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
34	124	F	169821	BDD	AAB	AAB	BCC	B	BBD	BBC	BCC	BBC	B	BDD	FCM
35	124	F	169822	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
36	126	F	169777	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	FCM
37	126	F	169778	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
38	126	F	169779	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
39	126	F	169780	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
40)	126	F	169781	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
41	126	F	169782	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
42	126	F	169783	BDD	AAB	ABB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
43	126	F	169784	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
44	126	F	169785	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
45	126	F	169786	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	FCM
46	126	F	169787	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
47	126	F	169788	BDD	AAB	ABB	BCC	B	BBD	BBD	AAC	BCC	B	BDD	FCM
48	126	F	169789	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
49	126	F	169790	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	FCM
50)	126	F	169791	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	FCM
51	126	F	169792	BDD	AAB	AAB	BCC	ABB	BBD	BBC	AAC	BCC	B	BDD	FCM

APPENDIX 2-5
(Continued)

	Site	Sex	AMNH	Locus											
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
52	126	F	169793	BDD	AAB	ABB	BCC	B	BBD	BBD	AAC	BBC	B	BDD	FCM
53	126	F	169794	BDD	AAB	AAB	BBC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
54	130	F	162938	BDD	AAB	QAA	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1027
55	130	F	165953	BDD	AAB	AAB	BCC	B	BBD	BBC	C	BCC	B	BDD	1032
56	130	F	165954	BDD	AAB	QAA	BCC	B	BBD	BBC	C	BCC	B	BDD	1115
57	130	F	165955	BDD	AAB	QAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1037
58	130	F	165956	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	907
59	130	F	165957	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	857
60)	131	F	165958	BDD	AAB	QAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1020
61	131	F	165959	BDD	AAB	AAB	BCC	B	BBD	—	C	BBC	B	BDD	996
62	131	F	165960	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	936
63	131	F	165961	BDD	AAB	ABB	BCC	B	BBD	—	C	BBC	B	BDD	841
64	131	F	165962	BDD	AAB	QAB	BCC	B	BBD	—	C	BCC	B	BDD	1042
65	137	F	167067	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1031
66	138	F	167071	BDD	AAB	—	BCC	B	BBD	BBC	—	B	B	BDD	1058
67	139	F	167072	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	996
68	140	F	169998	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
69	140	F	170000	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
70)	140	F	170001	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
71	141	F	170002	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
72	141	F	170003	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
73	141	F	170004	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
74	142	F	170005	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FMC
75	156	F	162925	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1147
76	156	F	162933	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1048
77	156	F	162934	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1035
78	156	F	162935	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1114
79	157	F	162926	BDD	AAB	AAB	BBC	B	BBD	BBC	—	BBC	B	BDD	—
80)	157	F	162927	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	1156
81	157	F	162928	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1385
82	157	F	162929	BDD	AAB	AAB	BCC	AAB	BBD	BBC	—	BBC	B	BDD	1143
83	157	F	162930	BDD	AAB	AAB	BBC	B	BBD	BBC	ABC	BCC	B	BDD	1084
84	157	F	162931	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1144
85	157	F	162932	BDD	AAB	AAB	BCC	B	BBD	—	ACC	BCC	B	BDD	1235
86	158	F	165963	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	976
87	158	F	165964	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	984
88	158	F	165965	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1043
89	158	F	165966	BDD	AAB	AAB	BBC	B	BDD	BCC	—	BBC	B	BDD	731
90)	158	F	165967	BDD	AAB	ABB	BBC	B	ABD	BBC	—	BBC	B	BDD	744
91	159	F	160322	BDD	AAB	AAB	C	B	BBD	BBC	ABC	BBC	B	BDD	965
92	159	F	160323	BDD	—	AAB	BCC	B	BBD	—	AAC	—	B	BDD	1000
93	159	F	160324	BDD	AAB	AAB	BCC	—	BBD	—	—	BBC	B	BDD	925
94	159	F	162922	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1031
95	159	F	162923	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1069
96	159	F	162924	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	989
97	160	F	165968	BDD	AAB	A	BCC	B	BBD	BBC	—	BCC	B	BDD	—
98	160	F	165969	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1129
99	160	F	165970	BDD	AAB	AAB	BCC	B	BBD	—	C	BCC	B	BDD	989
100)	160	F	165971	BDD	AAB	AAB	BBC	B	BBD	—	C	BCC	B	BDD	1086
101	161	F	166991	BDD	AAB	AAB	BCC	B	BBD	BBC	BBC	BCC	B	BDD	1133
102	161	F	166992	BDD	AAB	AAB	BCC	B	BBD	BBC	ABB	BCC	B	BDD	1397
103	161	F	166993	BDD	AAB	AAB	BBC	B	BBD	BBC	—	C	B	BDD	1142

APPENDIX 2-5
(Continued)

Locus															
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
104	161	F	166994	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	1084
105	161	F	166995	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1021
106	161	F	166996	BDD	AAB	ABB	BCC	B	BBD	BBC	BBC	BBC	B	BDD	1134
107	161	F	166997	BDD	AAB	A	BCC	B	BBD	BBC	—	BCC	B	BDD	1156
108	163	F	160329	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BCC	B	BDD	921
109	163	F	162890	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1232
110)	163	F	162891	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1223
111	163	F	162892	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1017
112	163	F	162893	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1084
113	163	F	162894	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	1128
114	163	F	162895	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1056
115	163	F	162896	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1064
116	163	F	162902	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	1094
117	164	F	162886	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	963
118	164	F	162897	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	1045
119	164	F	162898	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1070
120)	164	F	162899	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BCC	B	BDD	1173
121	164	F	162901	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1097
122	164	F	163554	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BCC	B	BDD	1080
123	165	F	160321	BDD	AAB	ABB	BBC	B	BBD	—	—	BCC	B	BDD	—
124	165	F	160325	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	1113
125	165	F	160326	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	1258
126	165	F	160327	BDD	AAB	AAB	BBC	B	BBD	—	AAC	BBC	B	BDD	950
127	165	F	160328	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	1347
128	165	F	162910	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BCC	B	BDD	1087
129	165	F	162911	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1065
130)	165	F	162912	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	1094
131	165	F	162913	BDD	ABB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1036
132	165	F	162914	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1021
133	165	F	162915	BDD	ABB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1069
134	165	F	162916	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	1132
135	165	F	162917	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	1108
136	165	F	162918	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1202
137	165	M	162919	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	1142
138	165	F	162920	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1204
139	165	F	162921	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1064
140)	166	F	162881	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1103
141	166	F	162882	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1091
142	166	F	162883	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	1111
143	166	F	162884	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BCC	B	BDD	1182
144	166	F	162885	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1195
145	166	F	162903	BBD	AAB	ABB	BCC	B	BBD	BBC	ABC	BBC	B	BDD	1148
146	166	F	162904	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BCC	B	BDD	1132
147	166	F	162905	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BBC	B	BDD	911
148	166	F	162906	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	BCC	B	BDD	1106
149	166	F	162907	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	1030
150)	166	F	162908	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BBC	B	BDD	989
151	166	F	162909	BDD	AAB	AAB	BCC	B	BBD	—	ABC	BBC	B	BDD	1113
152	167	F	162887	BDD	AAB	AAB	BCC	B	BBD	BCC	AAC	BBC	B	BDD	1209
153	167	F	162888	BDD	AAB	AAB	BCC	B	BBD	BCC	AAC	BBC	B	BDD	1022
154	167	F	162889	BDD	AAB	AAB	BCC	B	BBD	BCC	AAC	BBC	B	BDD	1021
155	169	F	167007	BDD	ABB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	1131

APPENDIX 2-5

(Continued)

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
156	169	F	167008	BDD	AAB	ABB	BCC	B	BBD	BCC	—	BCC	B	BDD	917
157	169	F	167009	BDD	AAB	AAB	BCC	B	BBD	BBC	BBC	C	B	BDD	1127
158	169	F	167010	BDD	AAB	A	BCC	B	BBD	BBC	—	BCC	B	BDD	1129
159	169	F	167011	BDD	AAB	AAB	BBC	B	BBD	BBC	BBC	C	B	BDD	1033
160)	179	F	169826	BDD	AAB	AAB	BCC	B	BBC	ABC	C	—	B	BDD	FCM
161	179	F	169827	BDD	AAB	AAB	BCC	B	BBD	AAB	—	BCC	B	BDD	FCM
162	179	F	169828	BDD	AAB	AAB	BCC	B	BBC	—	—	BCC	B	BDD	FCM
163	179	F	169829	BDD	AAB	A	BCC	B	BBC	ABC	C	—	B	BDD	FCM
164	179	F	169830	BDD	AAB	AAB	BCC	B	BBC	ABC	BCC	—	B	BDD	FCM
165	179	F	169831	BDD	AAB	AAB	BCC	B	BBC	—	—	BCC	B	BDD	—
166	179	F	169832	BDD	AAB	AAB	BCC	B	BBC	—	—	BCC	B	BDD	—
167	191	F	169839	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	FCM
168	191	F	162936	BDD	AAB	AAB	BCC	B	BBD	BBC	—	—	B	BDD	966
169	191	F	162937	BDD	AAB	AAB	BCC	B	BBD	BBC	—	—	BBC	BDD	1158
170)	191	F	162940	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	—	BDD	1104
171	191	F	162941	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	BCC	B	BDD	812
172	191	F	163458	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1009
173	192	F	169768	BDD	AAB	ABB	BCC	B	BBD	BBC	AAC	BBC	B	BDD	FCM
174	193	F	169769	BDD	AAB	AAB	BCC	B	BBD	BCC	—	B	B	BDD	FCM
175	193	F	169770	BDD	AAB	AAB	BCC	B	BBD	BCC	—	B	B	BDD	FCM
176	193	F	169771	BDD	AAB	AAB	BCC	BBC	BBD	BBC	—	B	B	BDD	FCM
177	194	F	165972	BDD	AAB	AAB	BCC	B	BBD	—	—	BBC	B	BDD	1106
178	194	F	165973	BDD	AAB	ABB	BCC	B	BBD	—	—	BBC	B	BDD	1031
179	194	F	165974	BDD	AAB	AAB	BCC	B	BBD	—	—	BBC	B	BDD	851
180)	194	F	165975	BDD	AAB	AAB	BCC	B	BBD	B	AAC	—	B	BDD	1029
181	194	F	165976	BDD	AAB	ABB	BCC	B	BBD	—	AAC	—	B	BDD	1060
182	194	F	165977	BDD	AAB	AAB	BCC	B	BBD	—	AAC	—	B	BDD	1142
183	194	F	165978	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	1211
184	195	F	158788	BDD	AAB	A	BCC	B	BBD	BBC	AAC	B	B	BDD	1022
185	195	F	158789	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1031
186	195	F	158790	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1000
187	195	F	158791	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1056
188	195	F	158792	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1010
189	195	F	158793	BDD	ABB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1033
190)	195	F	158794	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	966
191	195	F	158795	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	978
192	195	F	158796	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1003
193	198	F	158823	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	770
194	198	F	169349	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	FCM
195	198	F	169350	BDD	AAB	—	BCC	B	BBD	BBC	—	B	B	BDD	FCM
196	198	F	169351	BDD	AAB	—	BCC	ABB	BBD	BBC	—	B	B	BDD	FCM
197	200	F	169840	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
198	200	F	169841	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
199	200	F	169842	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
200)	200	F	169843	BDD	AAB	ABB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
201	200	F	169844	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
202	200	F	169845	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
203	200	F	169846	BDD	AAB	AAB	BCC	B	BBD	BBC	AAC	B	B	BDD	FCM
204	200	F	169847	BDD	AAB	AAB	BCC	B	BBD	BBC	A	B	B	BDD	FCM
205	200	F	169848	BDD	—	AAB	BCC	B	BBD	—	AAC	B	B	BDD	—
206	202	F	165979	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	932
207	202	F	165980	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1067

APPENDIX 2-5
(Continued)

Locus															
	Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood
208	202	F	165981	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1036
209	203	F	165982	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1029
210)	203	F	165983	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1081
211	203	F	165984	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1127
212	203	F	165985	BDD	AAB	AAB	BCC	B	BBD	—	AAC	BBC	B	BDD	1125
213	203	F	165986	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1076
214	203	F	165988	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	991
215	203	F	165989	BDD	AAB	ABB	BBC	B	BBD	BBC	—	B	B	BDD	1165
216	203	F	165990	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BBC	B	BDD	1070
217	203	F	165991	BDD	AAB	AAB	BCC	B	BBD	BBC	—	BCC	B	BDD	974
218	203	F	165992	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	1123
219	203	F	165993	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	1182
220)	207	F	169870	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
221	207	F	169871	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
222	207	F	169872	BDD	AAB	AAB	BCC	ABB	BBD	—	AAC	B	B	BDD	FCM
223	207	F	169873	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
224	207	F	169874	BDD	AAB	AAB	BCC	ABB	BBD	—	AAC	B	B	BDD	FCM
225	207	F	169875	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
226	207	F	169876	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
227	207	F	169877	BDD	AAB	AAB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
228	207	F	169878	BDD	AAB	AAB	BCC	ABB	BBD	—	AAC	B	B	BDD	FCM
229	207	F	169879	BDD	AAB	ABB	BCC	B	BBD	—	AAC	B	B	BDD	FCM
230)	208	F	158797	BDD	AAB	ABB	BCC	B	BBD	BBC	AAC	B	B	BDD	1356
231	208	F	158798	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1181
232	208	F	158799	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1140
233	208	F	158800	BDD	AAB	A	BBC	B	BBD	BBC	ABC	B	B	BDD	1144
234	207	F	158801	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1205
235	208	F	158802	BDD	AAB	A	BCC	B	BBD	BBC	AAC	B	B	BDD	1065
236	208	F	158803	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1146
237	208	F	158804	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1144
238	208	F	158805	BDD	ABB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1243
239	208	F	158806	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1077
240)	208	F	158807	BDD	AAB	AAB	BCC	B	BBD	BBC	ABC	B	B	BDD	1127
241	208	F	158808	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1168
242	208	F	158809	BDD	AAB	AAB	BCC	B	BBD	BBC	—	B	B	BDD	1116
243	208	F	158810	BDD	AAB	A	BCC	B	BBD	BBC	ABC	B	B	BDD	1089
244	213	F	153117	BDD	AAB	AAB	BCC	B	BBD	—	—	B	B	BDD	1020

Appendix 2-6: Genotypes of tetraploid (4n) *Ambystoma* (3) *laterale* – *jeffersonianum* (LLLJ) unisexual specimens ordered by site number.

	Site	Sex	AMNH	Locus											blood
				Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	
1	7	F	158825	BBBD	ABBB	ABBB	BBBC	B	BDDD	—	—	B	B	BBBD	1245
2	7	F	160339	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	BBCC	B	B	BBBD	1222
3	7	F	160340	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	BBCC	B	B	BBBD	1200
4	107	F	169899	BBBD	ABBB	ABBB	BBBC	A	BDDD	—	BCCC	B	B	BBBD	FCM
5	107	F	169909	BBBD	ABBB	ABBB	BBBC	B	BDDD	—	ABBB	B	B	BBBD	FCM
6	107	F	169910	BBBD	ABBB	ABBB	BBBC	B	BDDD	—	BCCC	B	B	BBBD	FCM
7	107	F	169911	BBBD	ABBB	ABBB	BBBC	B	BDDD	—	BCCC	B	B	BBBD	FCM
8	107	F	169912	BBBD	ABBB	ABBB	BBBC	B	BDDD	—	ABCC	B	B	BBBD	FCM
9	108	F	160341	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	BCCC	BBBC	B	BBBD	1212
10)	108	F	160342	BBBD	—	ABBB	BBBC	B	BDDD	—	—	BBBC	B	BBBD	1229
11	108	F	160343	BBBD	ABBB	ABBB	BBBC	B	BDDD	B	—	BBBC	B	BBBD	1286
12	110	F	169907	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	ACCC	B	B	BBBD	FCM
13	110	F	169908	BBBD	ABBB	B	B	B	BBDD	—	A	B	B	BBBD	FCM
14	111	F	169411	BBBD	ABBB	B	BBBC	B	BDDD	BCCC	ABCC	B	B	BBBD	FCM
15	111	F	169412	BBBD	ABBB	ABBB	BBBC	BBBC	BDDD	B	ACCC	B	B	B	FCM
16	111	F	169413	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	ACCC	B	B	BBBD	FCM
17	113	F	162950	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	AACC	B	B	BBBD	1317
18	148	M	167020	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	BCCC	B	B	BBBD	1190
19	155	F	160338	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	ACCC	B	B	BBBD	1251
20)	162	F	167004	BBDD	ABBB	AABB	BBCC	B	BDDD	BCCC	—	—	B	B	1133
21	168	F	160310	BBBD	ABBB	ABBB	BBBC	ABBB	BDDD	BCCC	—	BBBC	B	BBBD	1290
22	168	F	160318	BBBD	ABBB	ABBB	BBBC	ABBB	BDDD	—	ACCC	BBBC	B	BBBD	1030
23	168	F	160319	BBBD	ABBB	ABBB	BBBC	AABB	BDDD	—	ACCC	BBBC	B	BBBD	1113
24	198	F	169346	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	—	B	B	BBBD	FCM
25	198	F	169347	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	ACCC	B	B	BBBD	FCM
26	198	F	169348	BBBD	ABBB	ABBB	BBBC	B	BDDD	BCCC	ACCC	B	B	BBBD	FCM

Appendix 2-7: Genotypes of tetraploid (4n) *Ambystoma laterale* – (3) *jeffersonianum* (LJJJ) unisexual specimens ordered by site number.

Locus															
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
1	109	F	169922	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	ABCC	BCCC	B	BDDD	FCM
2	109	F	169923	BDDD	AAAB	AAAB	BCCC	ABBB	BBBD	—	ABCC	BCCC	B	BDDD	FCM
3	109	F	169924	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	ABCC	BCCC	B	BDDD	FCM
4	117	F	169456	BDDD	AAAB	A	BCCC	B	BBBD	BBBC	AAAC	BBBC	B	BDDD	FCM
5	117	F	169457	BDDD	AAAB	A	BCCC	B	BBBD	—	A	—	B	BDDD	FCM
6	117	F	169458	BDDD	AAAB	A	BCCC	B	BBBD	—	AAAC	—	B	BDDD	FCM
7	117	F	169459	BDDD	AAAB	A	BCCC	B	BBBD	BBBC	AAAC	BCCC	B	BDDD	FCM
8	126	F	169795	BDDD	AAAB	AAAB	BBBC	B	BBBD	BBBC	—	BBCC	B	BDDD	FCM
9	126	F	169796	BDDD	AAAB	AAAB	BCCC	B	BBBD	BBBC	—	BBBC	B	BDDD	FCM
10)	161	F	166998	BDDD	AAAB	AAAB	BCCC	B	BBBD	B	—	BCCC	B	BDDD	1396
11	179	F	169833	BDDD	AAAB	AAAB	BCCC	B	BBBD	BBBC	ABCC	C	B	BDDD	FCM
12	192	F	160377	BDDD	AAAB	AAAB	BCCC	B	BBBD	BBBC	AAAC	B	B	D	1536
13	194	F	166038	BDDD	AAAB	AAAB	BBBC	B	BBBD	BBBC	AAAC	BBBC	B	BDDD	1203
14	203	F	165987	BDDD	AAAB	AAAB	BCCC	B	BBBD	BBBC	—	B	B	BDDD	1089
15	203	F	166039	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	ACCC	B	B	BDDD	1447
16	203	F	166040	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	ACCC	B	B	BDDD	1332
17	203	F	166041	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	AAAC	B	B	BDDD	1317
18	203	F	166042	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	AAAC	B	B	BDDD	1387
19	207	F	169880	BDDD	AAAB	A	BCCC	B	BBBD	—	BBCC	B	B	BDDD	FCM
20)	207	F	169881	BDDD	AAAB	AAAB	BCCC	B	BBBD	—	AAAC	B	B	BDDD	FCM

Appendix 2-8: Genotypes of tetraploid (4n) *Ambystoma (2) laterale* – (2) *jeffersonianum* (LLJJ) unisexual specimen from site 18.

Locus															
Site	Sex	AMNH	Aat-1	Aat-2	Idh-1	Ldh-1	Ldh-2	Mdh-1	Mpi	Pgi	Pgm-1	Pgm-2	Sod-1	blood	
1	18	F	153121	BBDD	B	AABB	BBCC	B	BBDD	C	AACC	B	B	BBDD	—

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